

Improving Production of Cashew in Mozambique: A Preliminary Strategy

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STRATEGY**

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EXECUTIVE SUMMARY

Cashew has the potential to make a significant contribution to the economy of Mozambique. Increased cashew production can have a major impact on household income. However, it will take a concerted and coordinated national effort to address the problems that constrain cashew production, problems that have gone untreated since independence. If there is a consensus among the diverse and competing interests in the subsector, it is on the subject of production. Without a more abundant harvest of better quality nuts, farmers, processors and traders will not prosper.

Since May 1997, the USAID Mission to Mozambique has been providing technical assistance to the Cashew Working Group to assist the efforts of the subsector's principal stakeholders to develop a comprehensive rehabilitation strategy. The Mission contracted with the Agribusiness and Marketing Improvement Strategies (AMIS II) Project to do this. Over the course of the past several months, AMIS II consultants have examined a number of key production-related issues and begun to outline plans to address the most pressing constraints on production of more, higher quality nuts.

The strategy that is unfolding is sequential in nature. The first step is to improve the production from the existing national orchard. There is growing consensus that the production component of a national rehabilitation strategy needs to focus on identifying, propagating and distributing improved genetic material to increase production dramatically. But this is a medium- to longer-term proposition. In the short-term, the production strategy needs to focus on applying proven techniques to reduce the negative impact of major diseases and pests that constrain the ability of cashew trees in Mozambique to produce. The primary culprits are powdery mildew disease (*Oidium anacardii* Noack) and the sucking pest *Heliopeltis* spp.

In order to develop and propose a rational strategy to increase and improve production from existing trees, the AMIS II Project is currently preparing to undertake a series of crop protection trials. Dr. Clive Topper designed the trials program during a three week visit to Mozambique in the fall of 1997. Dr. Topper's original report forms the basis for the first two chapters of this document. In designing this program, the AMIS II Project has built upon the lessons learned recently in Tanzania where the crop protection experimentation has been extensive. The purpose here is to apply the scientific findings that are already available and thereby accelerate the implementation of measures to control disease and pests affecting cashew production in Mozambique.

The program includes both chemical and non-chemical approaches to combating powdery mildew disease (PMD). The chemical trials will take place under tightly controlled conditions at sites operated by two large private companies - Grupo Entrepasto and Grupo João Ferriera dos Santos - as well as the research station at Nassaruma, which is one of the major centers of operation for the African Development Bank-financed Cashew Rehabilitation Project. The non-

chemical or sanitary trials will measure the efficacy of pruning to reduce the impact of PMD. These will take place at several sites, some under the supervision of World Vision International and others under the Cashew Rehabilitation Project.

When the results of this series of trials are available in early 1999, the Cashew Working Group will have at least preliminary information with which to develop a broader program to reduce the negative impacts of the major diseases and pests affecting cashew production. For example, if the results of the sanitary trials prove favorable, there are a number of non-governmental organizations (NGOs) that might be mobilized to disseminate the mitigation techniques from the current effort. The Cashew Working Group would be the logical mechanism to mobilize support for and coordinate an extension effort to do this. That might require both the training of extension personnel affiliated with these organizations as well as the creation and reproduction of guides and other sources of information for farmers. Greater use of chemical control measures, should they prove successful, are likely to spread through the private sector firms which have the expertise to use these substances carefully and correctly.

Cashew production will not return to the level of the mid-1970s, when Mozambique was the world's largest producer, without a concerted effort to identify, propagate and distribute for planting improved genetic material. Although not part of his original mandate, Dr. Topper took it upon himself to produce a brief paper on how new techniques in breeding and multiplication might be applied to meet these longer term issues in Mozambique. His brief review comprises Chapter 3 of this document. It briefly reviews some of the recent efforts undertaken at Wye College in the U.K. in genetic fingerprinting, micropropagation and micrografting of cashew.

At the recommendation of Dr. Topper, Dr. Peter Caligari made a brief visit to Mozambique in December of 1997. His mandate was to review the current situation in Mozambique and begin to outline a strategy to improve cashew breeding. Chapter 4 of this report comprises Dr. Caligari's report and addresses the issue of how current scientific knowledge might be applied to address critical production constraints in Mozambique. Its key recommendation is that the Cashew Working Group host a workshop to define the ideotype for cashew trees to be exploited under different farming systems. It calls for a review and rationalization of cashew material (germplasm) in Mozambique.

When this report was originally distributed at the 10th Cashew Working Group meeting held in Nampula in February 1998, it was somewhat controversial. The above mentioned African Development Bank-financed Cashew Rehabilitation Project took exception to what it perceived to be an attack upon its approach to selecting "mother trees." Taken together, Dr. Caligari's original observations and Dr. Prasad's response, which is included as an Appendix to Chapter 4, begin to frame the debate about the direction in which a longer-term production strategy should evolve.

1. INTRODUCTION

Since gaining independence from Portugal in 1974, cashew production in Mozambique has declined precipitously. The growing prevalence of powdery mildew disease (PMD) and infestation have been primary factors for this decline. However, despite the prevalence of these problems, particularly in Nampula Province, which is the single most important cashew producing region in the country and an important focus of USAID-funded assistance programs in the country, the efforts currently underway to combat these major constraints on production are limited. Likewise, the whole research infrastructure of the country is weak and as a result there is not much research being undertaken to identify, propagate and disseminate improved genetic material is.

It is against this backdrop that the AMIS II project has begun to develop a framework for a strategy to rehabilitate cashew production in Mozambique. What is evolving is a two part strategy. In the short-term, the project hopes to identify short-term measures to improve the output of existing trees. Over the longer-term the project realizes that concerned stakeholders need to work together and pool their limited resources into a national effort to identify, propagate and disseminate improved genetic material.

1.1 Cashew Production in Mozambique

Cashew production in Mozambique peaked in 1972, when over 200,000 tonnes was marketed. After this, production fell off rapidly to less than 20,000 tonnes in 1982 and since then production has varied between 25,000 and 50,000 tonnes. During the period of peak production, Mozambique accounted for around 50 percent of the world market; recently it has accounted for less than 10 percent. The rise and fall of cashew production in Mozambique has remarkable similarities with that of neighboring Tanzania, where production peaked in 1974 at 145,000 tonnes and then declined dramatically to around 16,000 tonnes in 1986. However, since then production in Tanzania has more than quadrupled to reach 80,000 tonnes. There are an estimated 38 million cashew trees in Mozambique. Cashew was originally a major source of income for rural households, employment and foreign exchange. The cashew sub-sector will be instrumental in supporting agricultural-led growth in Mozambique.

In Nampula Province, the rainy season extends from December to April, with January, February and March being the high precipitation months. Local type trees flower mainly in June and July, while Brazilian dwarf trees tend to flower earlier in May. In the north, the main cashew harvesting period is between October and December, while in the south it occurs later between December and March. Nuts from the south tend to be of a lower quality compared with the north, and because production coincides with the Indian harvest, prices are lower. Nampula province, where all the trials will be located, is the main cashew producing area in Mozambique.

Apparently pigeon pea is extensively grown in Nampula province. The crop is normally harvested from May to July, which coincides with the main period of cashew flowering. This could have serious implications for cashew.

Over the last decade or more the cashew sub-sector has received funding from the African Development Bank (AfDB) and the World Bank. AfDB funding started in 1986/87, and up until 1994/95 progress was disappointing. Since then however, significant improvements have occurred. This project is due to be handed over to the GOM in 1998 and funding will end 1999.

The most obvious current cashew related activity in Mozambique is the replanting programme. The Cashew Rehabilitation Project is selling grafted seedling plants to small farmers for 3,000 meticaïs and to larger farmers for 5,000 meticaïs. Last season 70,000 grafted seedlings were sold and the target for the coming season is 250,000. The World Vision programme is also setting up nurseries to distribute grafted plants. Private companies have also become involved in the production and distribution of grafted plants. The majority of these plants will originate from locally selected trees.

The selection of local trees for the collection of scion to produce grafted plants for immediate distribution to farmers without prior controlled evaluation has been noted as a subject of concern by some experts. In addition to the genetical make-up of the tree, the environmental conditions within which a tree grows can have a great effect upon the yield produced.

Considering the importance of the cashew crop, the degree of cashew research currently being undertaken is quite low. This may be due to a lack of trained technical personnel, which also could have ramifications for undertaking trials in the future.

When the AMIS Project was carrying out field work in Nampula Province in the fall of 1997, the incidence of powdery mildew disease was widespread and clearly the main biotic constraint to cashew production. There were also areas where sucking pest (mainly *Helopeltis spp.*) damage was significant. This observation confirmed earlier quantitative surveys of cashew pest and disease problems that have been carried out in Nampula and other provinces, demonstrating that these problems still exist. (Rao and Nuvunga, 1993; Rao and Uaciquete, 1994; Nathaniels, 1994).

It is thought that mildew arrived or became a problem in Mozambique in the early 1980's. Trials to control cashew PMD were undertaken in 1993/94 by Mr. Jose Gilber Vasconcelos Lopes. The following fungicides were evaluated, Propriconazol (Tilt), Penconazol (Topas), Triadimenol (Bayfidan) + Chinometionato, Tebuconazol (Folicur) and Hexaconazol (Anvil). The dose rate/tree was difficult to determine, method of application was by manual knapsack sprayer. The final report states that the "data is being analyzed statistically" but in a personal communication to Dr Prasad, it was indicated that the results were inconclusive.

Mr. Paulo R. De Carvalho undertook one trial to control PMD and sucking pests using the following insecticides: Cyfluthrin (Baythroid), Beta-Cyfluthrin (Bulldock) and Imidacloprid (Confidor) and the fungicide Tebuconazole (Folicur) which was used alone and in conjunction with each of the insecticides. Treatments without Folicur had an average level of PMD of 99.25

percent while those with Folicur resulted in an average of 76 percent. It would appear that Folicur did reduce the PMD infection but not significantly. Treatments without insecticide had an average *Helopeltis* infestation of 30.5 percent, while insecticide treatments had much lower levels at an average of 10.9 percent.

It appears that no other crop protection trials on cashew have been undertaken, at least in Nampula Province.

1.2 Purpose and Structure of This Report

The production component may be the single most important part of the AMIS II Project's broader mandate to work with local stakeholders to develop a rehabilitation strategy for the entire cashew subsector in Mozambique. This report comprises an initial analysis to identify options and outline both short-term strategies to improve increase production from existing trees and longer term strategies to improve the quantity and quality output with improved genetic material.

This report differs in several important ways from typical AMIS II Project deliverables. It is a consolidation of three separate reports prepared by two different AMIS II consultants over the course of the past several months. In the fall of 1997, Dr. Clive Topper undertook a consultancy to study what might be done in the near term to alleviate production constraints. His report provides an overview of the current situation in Nampula Province, the single most important cashew growing region of the country. In addition, it presents a proposal for a series of crop protection trials to test different approaches to mitigate and control the negative effects of powdery mildew disease (PMD) and infestation. Chapter 2 of this document is an edited version of Dr. Topper's proposal.

Chapters 3 and 4 begin to address the issue of the options Mozambique has to develop a longer-term production rehabilitation strategy. The focus here is to develop a national strategy to select the most appropriate genetic material available to reconstitute the national orchard with trees that will be tolerant of , if not resistant to, PMD. Dr. Topper prepared the report which is the basis of Chapter 3 at the end of his initial consultancy. His recommendation for a follow-up consultancy resulted in a trip by Dr. Peter Caligari in December 1997. Chapter 4 is an edited version of Dr. Caligari's report.

The appendices to each chapter include relevant background material, summaries of site visits, and bibliographies.

2. PROPOSAL FOR A SERIES OF CROP PROTECTION TRIALS

Powdery mildew disease (*Oidium anacardii* Noack) is widespread in Nampula Province and is the main biotic constraint to cashew production. There are also areas where sucking pest (mainly *Helopeltis* spp.) damage is significant. To develop a strategy to mitigate these problems and improve cashew production, the AMIS II Project engaged the services of Dr. Clive Topper to design a series of crop protection trials. His terms of reference called for him to design trials to test both chemical and non-chemical (sanitary) approaches to control and mitigate the negative impacts of powdery mildew disease (PMD), as well as to test the efficacy of using predatory ants to control *Helopeltis* spp. The proposed program of trials presented in this chapter is the final product of Dr. Topper's consultancy.

2.1 Introduction to Chapter 2

In the fall of 1997 AMIS II consultant Dr. Clive Topper visited Mozambique to gather preliminary information and meet with representatives of government organizations, private companies and NGOs who might collaborate with the project on this undertaking. Given his intimate knowledge of the extensive work to control disease and infestation that has been carried out recently in Tanzania, Dr. Topper was particularly well qualified for this assignment. (Appendix 1 to this chapter provides a brief summary of the Tanzanian experience.)

This chapter sets forth a comprehensive plan for trials to control powdery mildew disease (PMD) in the coming cashew season. The proposed sites for chemical control trials are Nassuruma Research Station, Entrepoto plantation at Monapo and the JFS plantation at Geba. Appendix 2 provides a summary of the visits Dr. Topper made to each of the proposed trial sites. Trials will be undertaken both on local, normal size trees and Brazilian dwarf trees. The plan also includes a proposal to carry out a cultural control programme (phytosanitation) against powdery mildew in collaboration with the Cashew Rehabilitation Project and World Vision. Another trial planned with World Vision will attempt to define the factors responsible for low yields from trees seemingly unaffected by mildew. Preliminary work to establish the role of the major insect pest (*Helopeltis*) and the extent of its biological control by the predacious ant, *Oecophylla*, is proposed.

All trials will provide data on the effects of PMD and *Helopeltis* on yield, regardless of whether the trial is specifically aimed at PMD control or *Helopeltis* control. Quantitative data on flowering cycles will also be obtained from all trials, including the relative importance of the second flowering cycle that occurs in September/October.

The chapter includes a comprehensive list of equipment and pesticides required for the proposed trials along with detailed costings. a programme of visits and the work to be done at the various trial sites is given for different times during the cashew season.

Finally, readers concerned with the environmental aspects of the proposed trials and with meeting USAID requirements on pesticide use and safety, should refer to the separate report prepared by Dr. E.A. Heinrichs¹.

2.2 Summaries of Proposed Trials

The paragraphs below summarize different trials the AMIS II Project proposes to conduct at six (6) different sites.

2.2.1 Trial 1: Chemical Control of PMD - Nassuruma site

Collaborating institution: Cashew Rehabilitation Project (CRP)

Objective:

To evaluate the effectiveness of 3 fungicides and 2 dose rates for the control of PMD.

Background:

The proposed block has been abandoned for a few years and is in need of a complete clean up, the estimated cost is about \$1,500. Apparently yield records do exist for these trees for some years, and these data could be of use when analyzing the chemical control trial results. One of the technicians, Mr. Consolo, has had crop protection training during a 6-month visit to Brazil. *Oecophylla* was present on some trees.

Treatments:

Complete randomized block factorial trial with the following factors:

- a. Fungicide (Anvil, Bayfidan and Sulphur)
- B. Dose rate (low and high)

Check or control plots will also be included

Details of quantities involved:

Factor a	Fungicide	Anvil	Anvil	Bayfida	Bayfida	Sulphur	Sulphur	
Factor B	Dose rate	10	15	10	15	170	250	Total no. of plots

¹ Heinrichs, E.A. 1997. *Development of Pest Management Strategies for Cashew in Mozambique*. Blacksburg, VA: Integrated Pest Management Program Collaborative Research Support Program (IPM CRSP).

Replicate	4	4	4	4	4	4	24
Plot size (no. of trees)	50	50	50	50	50	50	Total no. of trees
Total no. trees/treat =	200	200	200	200	200	200	1200
No. of applications	4	4	4	4	4	4	
Qty of chemical ml or	8000	12000	8000	12000	136000	200000	
Total qty of each chemical		20 litres		20 litres		336 kg	

Proposed general work programme :

Prior to April - the site needs to be thoroughly cleaned and fire breaks completed

May - identify trees with *Oecophylla* present

June - set-up chemical control trial and mark out plots

June, July - evaluate mildew, *Helopeltis* damage and flowering

August, September - detailed PMD score and continue monitoring *Helopeltis* damage

October to December - yield data collection

Sampling programme : (to start with panicle emergence)

Monday Replicate 1, 7 plots x 10 trees/plot = 70 trees

Tuesday Replicate 2, 7 plots x 10 trees/plot = 70 trees

Wednesday Replicate 3, 7 plots x 10 trees/plot = 70 trees

Thursday Replicate 4, 7 plots x 10 trees/plot = 70 trees

Friday *Oecophylla* sampling

NB: the same person should sample Complete replicates every time.

CRP commitment:

Allow trees to be used for trial.

Clean up block of trees and complete fire breaks

Provide 2 full time technicians and transport to allow them easy field access

Provide supervisor to periodically check that the work is being carried out as planned

Provide a pick-up vehicle and 2 laborers for spraying (probably for a total of 20 days)

USAID Project commitment:

Provide training for the technicians

Collate, analyze and report on data collected

Provide results to CRP and other appropriate parties

Provide the following equipment and chemicals for trial use:

Items produced outside of Mozambique (but may be available locally):

Sprayers/dusters and spares	3
Fungicides	
Anvil (litres)	20
Bayfidan (litres)	20
Sulphur (kg)	336
Measuring cylinders	
250 ml	1
500 ml	1
1 litre	1
Protective clothing	2
Masks	10
Gloves	4
Respirator	2
Compass	1
Balance	2

Items to be purchased locally:

Sieve	1
Petrol container	1
2 stroke oil container	1
Water containers 25 litres	10
Funnel (petrol)	1
Pencils	20
Pencil sharpeners	2
Clip boards	3
Data sheets	

2.2.2 Trial 2: Chemical control of PMD and enhancement of *Oecophylla* - Geba site

Collaborating institution: Grupo Joao Ferreira dos Santos (JFS)

Objective:

1. To evaluate the effectiveness of 3 fungicides for the control of PMD in the presence/absence of *Oecophylla*
2. To enhance the existing population of *Oecophylla* and increase their territory.

Background:

Geba is located across the bay from Nacala, approx. 80km from the main road and 2 hours drive from Namialo (JFS headquarters in Nampula province). There is a block of about 80 ha of older trees. The site is particularly suitable because of the proportion of trees colonized by *Oecophylla* and the difference in sucking pest damage between those colonized and those not.

Treatments for Objective 1:

Complete randomized block factorial trial with the following factors:

- a. Fungicide (Anvil, Bayfidan and Sulphur)
- B. Dose rate (low and high)
- C. *Oecophylla* (present and absent)

Details of quantities involved:

Factor a	Fungicide	Anvil	Anvil	Bayfida	Bayfida	Sulphur	Sulphur	
Factor B	Dose rate	10	15	10	15	170	250	Total no. of plots
Replicate		4	4	4	4	4	4	24
Plot size (no. of trees)		50	50	50	50	50	50	Total no. of trees
Total no. trees/treat =		200	200	200	200	200	200	1200
No. of applications		4	4	4	4	4	4	
Qty of chemical		8000	12000	8000	12000	136000	200000	
Total qty of each chemical			20 litres		20 litres		336 kg	

Treatments for Objective 2:

Introduction of new colonies

Aerial walkways

Proposed general work programme :

Prior to April - the site needs to be thoroughly cleaned (this is already part of the JFS programme).

March – the area needs to be mapped and each tree evaluated for presence/absence of *Oecophylla*

April/May - introduce new colonies and erect walkways

June - set-up chemical control trial and mark out plots

June, July - evaluate mildew, *Helopeltis* damage and flowering

August, September - detailed PMD score and continue monitoring *Helopeltis* damage

October to December - yield data collection

JFS commitment:

Allow trees to be used for trial.

Clean up block of trees (already in JFS programme)

Provide 2 full time technicians and transport to allow them easy field access

Provide a pick-up vehicle and 2 laborers for spraying (probably for a total of 20 days)

Provide supervisor to periodically check that the work is being carried out as planned

Guest house facilities

USAID Project commitment:

Provide training for the technicians

Collate, analyze and report on data collected

Provide results to JFS and other appropriate parties

Provide the following equipment and chemicals for trial use:

Items produced outside of Mozambique (but may be available locally):

Sprayers/dusters and spares	3
Fungicides	
Anvil (litres)	20
Bayfidan (litres)	20
Sulphur (kg)	336
Measuring cylinders	
250 ml	1
500 ml	1
1 litre	1
Protective clothing	2
Masks	10
Gloves	4
Respirator	2
Compass	1
Balance	2

Items to be purchased locally:

Sieve	1
Petrol container	1
2 stroke oil container	1
Water containers 25 litres	10
Funnel (petrol)	1
Pencils	20
Pencil sharpeners	2
Clip boards	3
Data sheets	

2.2.3 Trial 3: Chemical control of PMD on (a) 8 year old local trees and (B) on Brazilian dwarf trees - Monapo site

Collaborating institution: Entrepoto

Objective:

To evaluate the effectiveness of 3 fungicides and 2 dose rates for the control of PMD on both non-selected local trees and Brazilian dwarf trees.

Introduction:

Block of 8 year old trees – this block runs either side of a main road and in spite of the efforts of guards, some of the yield still disappears, therefore the trial plots should be positioned with this in mind. Individual plots should be 3 rows wide and about 11 trees long, in this way one whole plot can be sprayed with 1 full tank of the motorized blower. Sampling would be confined to the five central trees of the middle row.

X X X X X X X X X X X

X X X **X X X X X** X X X

X X X X X X X X X X X

Block of 3-year-old Brazilian dwarf trees – the plot layout should be the same as above.

Treatments:

Complete randomized block factorial trial with the following factors:

- a. Fungicide (Anvil, Bayfida and Sulphur)
- B. Dose rate (low and high)

No. of replicates = 6 on **both local and Brazilian trees**

Details of quantities involved – 8 year old local trees:

Factor a	Fungicide	Anvil	Anvil	Bayfida	Bayfida	Sulphur	Sulphur	
Factor B	Dose rate	5	10	5	10	85	170	Total no. of plots
Replicate		6	6	6	6	6	6	36
Plot size (no. of trees)		30	30	30	30	30	30	Total no. of trees
Total no. trees/treat =		180	180	180	180	180	180	1080
No. of applications		4	4	4	4	4	4	
Qty of chemical		3600	7200	3600	7200	61200	122400	
Total qty of each chemical			10.8 litres		10.8 litres		183.6 kg	

Details of quantities involved – Brazilian dwarf trees:

Factor a	Fungicide	Anvil	Anvil	Bayfida	Bayfida	Sulphur	Sulphur	
Factor B	Dose rate	2.5	5	2.5	5	42.5	85	Total no. of plots
Replicate		6	6	6	6	6	6	36
Plot size (no. of trees)		50	50	50	50	50	50	Total no. of trees
Total no. trees/treat =		300	300	300	300	300	300	1800
No. of applications		4	4	4	4	4	4	
Qty of chemical		3000	6000	3000	6000	51000	102000	
Total qty of each chemical			9 litres		9 litres		153 kg	

Proposed general work programme :

Prior to April - the site needs to be thoroughly cleaned

May - set-up chemical control trial and mark out plots

May, June, July - evaluate mildew, *Helopeltis* damage and flowering

August, September - detailed PMD score and continue monitoring *Helopeltis* damage

October to December - yield data collection

Entreposto commitment:

Allow trees to be used for trial.

Clean up block of trees

Provide 2 full time technicians and transport to allow them easy field access

Provide supervisor to periodically check that the work is being carried out as planned

Provide a pick-up vehicle and 2 laborers for spraying (probably for a total of 25 days)

Guest house facilities

USAID Project commitment:

Provide training for the technicians

Collate, analyze and report on data collected

Provide results to Entreposto and other appropriate parties

Provide the following equipment and chemicals for trial use:

Items produced outside of Mozambique (but may be available locally):

Sprayers/dusters and spares	3
Fungicides	
Anvil (litres)	20
Bayfidan (litres)	20

Sulphur (kg)	340
Measuring cylinders	
250 ml	1
500 ml	1
1 litre	1
Protective clothing	2
Masks	20
Gloves	4
Respirator	2
Compass	1
Balance	2

Items to be purchased locally:

Sieve	1
Petrol container	1
2 stroke oil container	1
Water containers 25 litres	10
Funnel (petrol)	1
Pencils	20
Pencil sharpeners	2
Clip boards	3
Data sheets	

2.2.4 Trial 4: Cultural control of PMD (phytosanitation) - numerous sites

Collaborating institution: 1. Cashew Rehabilitation Project (CRP)
2. World Vision

Objective:

To delay the start and intensity of the powdery mildew epidemic in order to allow a greater proportion of flowers from the first flowering cycle to mature and set nuts.

Proposed sites: 20 villages distributed in the districts to the south of Nampula (CRP)
2 sites in districts to the north of Nampula (World Vision)

Background:

The actual activity of sanitizing the trees has to fit in with the farmers' other commitments; i.e. planting and maintenance of food crops. Possible problems include some farmers within the sanitized block who are unwilling or unable to sanitize their trees and also the presence of ownerless trees.

Treatments:

1. Removal of all green shoots under the canopy for a period of 3 to 4 months prior to the start of panicle emergence (see diagramme below).
2. “Check” area in which no PMD control measures are undertaken

The sanitation area should be composed of a minimum of 10 farmers (each farmer owns an average of 60 trees) but preferably, 15 to 20 farmers, this would give between 900 and 1200 trees in the sanitation area. If possible the sanitation area should be bordered by an area devoid of cashew.

Proposed general work programme :

October 1997 - define sanitation and check areas. Collect information from farmers in both the sanitation and check areas regarding the yield from the **1997 first flowering cycle** (i.e. harvested during September, October and November). This should take the form of a table, one each for the sanitized area and the check areas, e.g.

	Farmer name	Total no. of trees	Total yield in 1997 from 1st flowering cycle
1			
2			
3			
4			

January/February - first round of sanitation

March/April - second round of sanitation

April/May - third round of sanitation

June and July - PMD scoring

September - Detailed PMD score

September to November - yield data collection

CRP commitment:

To provide technicians (20 in number) to train the farmers in the technique of sanitation and to spend 1 day per week from May/June to end of July monitoring PMD and flowering patterns, and then approx. 1 day per 2 weeks monitoring PMD and yield.

Provide supervisor to periodically check that the work is being carried out as planned.

World Vision commitment:

To provide technicians (2 in number) to train the farmers in the technique of sanitation and to spend 1 day per week from May/June to end of July monitoring PMD and flowering patterns, and then approx. 1 day per 2 weeks monitoring PMD and yield.

Provide supervisor to periodically check that the work is being carried out as planned.

USAID Project commitment:

To provide field training on the technique of tree sanitation.

To provide training on the scoring of PMD, flowering cycle evaluation and yield data collection to evaluate the success of the technique.

To assess the extent and the quality of implementation of the first round of sanitation and again prior to panicle emergence.

To provide 1 machete to each farmer involved.

To collate, analyze and report on data collected.

Distribute report to CRP and other appropriate parties.

2.2.5 Trial 5: Determine factors causing negligible yields from trees NOT affected by Powdery Mildew

Collaborating institution: World Vision

Objective:

a large proportion of the cashew tree population produces very low yields but appears **NOT** to be significantly affected by powdery mildew disease. Thus, some other factor(s) is responsible. This study aims to determine the factors involved.

Background:

The main characteristic of these trees is that the panicles appear long and thin, with few lateral branches and of a black colour. The few flowers that remain by October (perhaps only a few are produced) have developed normally but have then dried out to give a brown colour (this is in contrast with mildew affected flowers which tend to be grey/black in colour). Often the leaves of the trees have an anemic colour (lime green) with reduced leaf density. These trees tend to be located away from the main thoroughfares; i.e. villages and main roads, where, in contrast, the trees tend to be the normal dark green with a dense leaf canopy.

Proposed general work programme :

No. of technicians involved: 9

No. of trees to be sampled by each technician: 50

Total no. of trees to be sampled: 450

October 1997 - technicians select 50 trees each with the above characteristics, mark the trees with a painted number (1-50) (the crushed end of a twig can be used as a paintbrush) and draw a sketch map of the area so as to enable easy access. The trees should be spread out but not too distant so as to make sampling too time consuming.

May, June and July - during this time the following parameters should be sampled weekly:

No. of shoots

No. of flushing shoots

No of emerging panicles
Panicle size, no. of branches, no. of flowers
Presence or absence of PMD
No. of black lesions (*Helopeltis* feeding sites)
No. of young and mature nuts

August, September and October - visual follow-up of panicle development.

World Vision commitment:

To spend 1 or 2 days in October 1997 (i.e. as soon as possible) to identify experimental trees.
To allow all technicians to attend 2 day training workshop in May.
To provide from mid May to July, 1 day per week from each technician (9) and from August to October, ½ a day per 2 weeks from each technician.
Provide supervisor to periodically check that the work is being carried out as planned.

USAID Project commitment:

Provide 2 day training workshop for the technicians
Provide the following equipment for trial use
Data forms (approx. 25 per technician)
< Pencils
< Clip boards.

To collate, analyze and report on data collected
Provide results to World Vision and other appropriate parties

If the factors responsible for the low yields are not obvious from this study, it is suggested that soil and leaf samples are taken for analysis (this activity has not been costed).

2.2.6 Trial 6: Trunk injection trial – 2 sites

Objective:

To evaluate the effectiveness of systemic fungicides injected into the trunk of the tree for the control of powdery mildew disease.

Background:

Fungicides are normally applied to tree crops as foliar sprays (or soil drenches). The effectiveness of spraying methods, as protective or curative treatments for plant diseases, is limited by the inefficiency of the spraying system, by the uptake into the plant tissue, by degradation on the leaf surface and loss to the environment by wash off. Trunk injection is a novel technique that delivers a low volume of fungicide into the tree with minimum wastage and environmental contamination and achieves maximum persistence within the plant.

The objective will be to find systemic fungicides that are effective against powdery mildew diseases **and** can be translocated in the xylem or cambial zone, from the site of injection to the leaves and panicles (i.e. in the transpiration stream to the periphery of the tree). Should the technique work, the advantage would lie mainly in reducing application costs.

Treatments for PMD control:

Complete randomized block factorial with the following factors:

- a. Fungicide (Triadimenol, Propiconazole and Myclobutanil)
- B. Dose rate/injection site (high and low)

Treatments for insect control:

Complete randomized block factorial with the following factors:

- a. Insecticide (Imidacloprid and Abamectin)
- B. Dose rate/injection site (high and low)

It should be noted that the manufacturers of the Wedgle injection system recommend the use of specially formulated pesticides which are extremely expensive (see Section 4.1). Injections should be made at 10 to 15cm intervals around the circumference of the tree. The Table below gives the range in injection sites for 3 different size trees.

Trunk diameter	Circumference	No. of injections at intervals of:	
		10cm	15cm
20cm (8")	62.8cm	6	4
30cm (12")	94.3cm	9	6
45cm (18")	141.4cm	14	9

Taking a typical recommended dose rate of 1ml/injection site and a typical tree of 30cm diameter trunk, the minimum amount of fungicide or insecticide required would be 6ml. The cost of 6ml would be \$12 for chemical alone. Compare this with the cost of conventional fungicide spraying, where the cost of chemicals to treat one large tree is less than \$2. Obviously it will be necessary to determine the absolute minimum dose rate commensurate with good control. Also the injector should be tried with normal pesticide formulations, which are much cheaper.

Plot size = 2 medium sized trees

No. of replicates = 4

No. of sites = 2 (two sites from either Geba, Monapo or Nassuruma, to be decided at the time)

2.3 Summary of Equipment and Pesticide Quantities and Cost

The paragraphs below provide a summary of the quantities and costs of the equipment and pesticides that the program of trials described above will require.

2.3.1 Pesticide Costs

Trade name	Common Name	Formulation	Active ingredient	Cost	Source
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Pesticides for spray application

Anvil	Hexaconazole	SC5	50g ai/litre	\$25/l	Zeneca
Bayfidan	Triadimenol	EC250	250g ai/litre	\$30/l	Bayer
Karate	Lamda cyhalothrin	EC50	50g ai/litre	\$12/l	Zeneca

The above pesticides are readily available in Mozambique.

Pesticides for trunk injection – PMD control

Bayfidan	Triadimenol	TI *	120ml bottle	\$250	Arborsystems
Tilt or Alamo	Propiconazole	TI *	120ml bottle	\$250	Arborsystems
Systhane	Myclobutanil	TI *	120ml bottle	\$250	Arborsystems

Pesticides for trunk injection – insect control

Confidor	Imidacloprid	TI *	120ml bottle	\$250	Arborsystems (Bayer)
Avid	Abamectin	TI *	120ml bottle	\$250	Arborsystems

* TI – special formulation for Trunk Injection; without it, the suppliers of the injection system maintain that it will not work, hence the high cost (prices obtained over the telephone).

2.3.2 Motorised Blower Costs

Solo Port 423 motorised blower, 70cc engine (spray tank = 12 litres) *¹
\$881.70

Solo Port 423 motorised blower, 70cc engine (spray tank = 12 litres) *² PS 467.23 or
\$747.57

Allman L80A motorised blower, 77cc engine (spray tank = 14 litres) *² PS 379.00 or
\$606.40

- *1 Cost in Mozambique
 *2 Cost in UK, shipping costs to be added.

The most widely used blower in Tanzania is the Maruyama (price to be determined).

(Approx. exchange rate used, Pound Sterling (PS) 1 = \$1.6)

2.3.3 Issues regarding equipment, procurement and budgets to be resolved in near future

a few things still need to be resolved regarding equipment, procurement and budgets:

Who will be responsible for the equipment in Mozambique?

- C Ownership of equipment after trials (preferably it all should be kept and well maintained in case required for trials in 1999).
- C Where should the equipment be consolidated prior to distribution to trial sites (and stored after the trials).
- C All equipment should be in country by March, ready for testing and training, therefore decisions about budgets, procurement, etc. need to be made fairly soon.
- C a small budget should be available in-country for small items required at the time of the trials and an account holder should be appointed.

It is suggested that the consultant coordinate the purchase of all the equipment outside of Mozambique and that the Cashew Working Group Secretariat/GOM scientist make arrangements for purchases within Mozambique.

2.3.4 Quantity and costs of equipment and pesticides required

Table 1. Quantity and costs of equipment and pesticides required specifically for trunk injection trial

Item	Quantity	Unit Cost \$	Total cost \$
Wedge system	3	375	1,125
Wedge spare needles	12	5	60
Bayfidan	120ml bottle	250	250
Tilt/Alamo	120ml bottle	250	250
Systhane	120ml bottle	250	250
Confidor/Merit	120ml bottle	250	250
Avid	120ml bottle	250	250
TOTAL			2,435

Table 2. Quantity and costs of equipment and pesticides required for remaining trials

a. Items produced outside of Mozambique (but may be available locally)

	Sanitati CRP	Sanitati W	Low W	Chem Nassuru	Chem 8 Manap	Braz Manapo	Chem/a Geba	TOT	UNIT COS	TOT COS
Sprayers/duster				3	3	-	3	9	800	7,200
Fungicides				-	-	-	-			
Anvil				20	11	9	20	60	25	1,500
Bayfidan				20	11	9	20	60	30	1,800
Sulphur (kg)				340	190	160	340	1030	2	2,060
Insecticide								10	12	120
Measuring				-	-	-	-			
25 ml				2	2			4	4.6	18
50 ml				2	2			4	4.8	19
250 ml				1	1	1	1	4	10.5	42
500 ml				1	1	1	1	4	15.8	63
1 litre				1	1	1	1	4	22.5	90
Protective				2	2	1	2	7	30	210
Masks				10	10	10	10	40	1.5	60
Gloves				4	4	4	4	16	3.5	56
Respirator				2	2	1	2	7	18	126
Respirator				8	8	4	8	28	6	168
Goggles				2	2	1	2	7	13.5	95
Compass				1	1	-	1	3	10	30
Balance				2	2	1	2	7	30	210

b. Items available locally

Sieve				1	1	1	1	4	2	8
Petrol				1	1	-	1	3	25	75
2 stroke oil				1	1	-	1	3	2	6
Water				10	10	-	10	30	15	450
Funnel (petrol)				1	1	-	1	3	3	9
Pencils	60	10	30	10	10	10	10	140	0.3	42
Pencil	20	2	9	2	2	-	2	37	1	37
Clip boards	20	2	9	2	2	-	2	37	10	370
Data sheets										
Machete	300	100						400	3.5	1,400

TOTAL COST 16,26

a total equipment budget, including contingency and local operating budget should therefore amount to about \$22,000. The GOM scientist should have unrestricted access to a computer, therefore it might be necessary to purchase a laptop computer.

2.4 Proposed programme of visits by consultants for trial implementation

Table 3, below, provides a summary calendar for carrying out the proposed program of trials.

Table 3. Timetable of proposed visits by consultants (D = Detailed scoring, Y = Yield)

		No of days	Sanitation	Sanitation	Low yields	Chem trial	Chem 8 yr	Braz dwarfs	Chem/ants
Date		General	CRP	W Vision	W Vision	Nassuruma	Monapo	Monapo	Geba
12-Jan	Mon	1	Prepare equipment						
13-Jan	Tue	2	Prepare equipment						
14-Jan	Wed	3	Prepare equipment						
19-Jan	Mon	1	Prepare manuals						
20-Jan	Tue	2	Prepare manuals						
21-Jan	Wed	3	Prepare manuals						
22-Jan	Thu	4	Prepare manuals						
23-Jan	Fri	5	Prepare manuals						
02-Feb	Mon	1	Depart						
03-Feb	Tue	2	Arrive Maputo						
04-Feb	Wed	3	Arrive Nampula						
05-Feb	Thu	4	Check						
06-Feb	Fri	5	Check						
07-Feb	SAT	6	Training						
08-Feb	SUN	7	Training	Check					
09-Feb	Mon	8	Data 97	Training					
10-Feb	Tue	9		Data 97	Check				
11-Feb	Wed	10				Check map			
12-Feb	Thu	11				Ants/yield			
13-Feb	Fri	12	Equipment						
14-Feb	SAT	13	Arrive Maputo						
15-Feb	SUN	14	Depart Maputo						
16-Feb	Mon	15	Arrive						
20-Apr	Mon	1	Depart						
21-Apr	Tue	2	Arrive Maputo						
22-Apr	Wed	3	Arrive Nampula						
23-Apr	Thu	4	Check						
24-Apr	Fri	5	Check						
25-Apr	SAT	6	Train scouting						
26-Apr	SU	7	Train scouting						
27-Apr	Mon	8		Check					
28-Apr	Tue	9		Train	Train scouting				
29-Apr	We	1				Check			
30-Apr	Thu	1				Plot layout		Plot layout	
01-May	Fri	1						Train scouting	
02-May	SAT	1						Train	Plot layout
03-May	SU	1							Ants
04-May	Mon	1	Check equipment						
05-May	Tue	1	Arrive Maputo						
06-May	We	1	Depart Maputo						
07-May	Thu	1	Arrive UK						
		8							

Continued

		No of days	Sanitation	Sanitation	Low yields	Chem trial	Chem 8 yr	Braz dwarfs	Chem/ants
Date		General	CRP	W Vision	W Vision	Nassuruma	Monapo	Monapo	Geba
08-Jun	Mon	1	Depart UK						
09-Jun	Tue	2	Arrive Maputo						
10-Jun	We d	3	Arrive Nampula						
11-Jun	Thu	4						Training	
12-Jun	Fri	5					Plot layout	Training	
13-Jun	SAT	6					Plot layout		
14-Jun	SU N	7					Train scout		
15-Jun	Mon	8					Train blower		
16-Jun	Tue	9							Plot layout
17-Jun	We d	10							Plot layout
18-Jun	Thu	11							Train scout
19-Jun	Fri	12							Train blower
20-Jun	SAT	13				Plot layout			
21-Jun	SU N	14				Plot layout			
22-Jun	Mon	15				Train scout			
23-Jun	Tue	16				Train blower			
24-Jun	We d	17	Check/train						
25-Jun	Thu	18	Check/train						
26-Jun	Fri	19	Check/train						
27-Jun	SAT	20		Check/train	Check/train				
28-Jun	SU N	21		Check/train	Check/train				

29-Jun	Mon	2	Arrive Maputo						
		2							
30-Jun	Tue	2	Depart Maputo						
		3							
01-Jul	We d	2	Arrive						
		4	UK						
10-Aug	Mon	1	Depart UK						
11-Aug	Tue	2	Arrive Maputo						
12-Aug	We d	3	Arrive Nampula						
13-Aug	Thu	4		Check					
14-Aug	Fri	5		Train D & Y					
15-Aug	SAT	6			Check				
16-Aug	SU N	7			Train D & Y	Check			
17-Aug	Mon	8				Train D & Y	Check		
18-Aug	Tue	9					Train D & Y		
19-Aug	We d	1					Check	Check	
		0							
20-Aug	Thu	1					Train D &Y	Train D & Y	
		1							
21-Aug	Fri	1							Check
		2							
22-Aug	SAT	1							Train D & Y
		3							
23-Aug	SU N	1	Arrive Maputo						
		4							
24-Aug	Mon	1	Depart Maputo						
		5							
25-Aug	Tue	1	Arrive						
		6	UK						
14th September to 10th October (24 days) - Analysis of flowering, PMD and Helopeltis damage									
02-Nov	Mon	1	Depart UK						
03-Nov	Tue	2	Arrive Maputo						
04-Nov	We d	3	Arrive Nampula						
05-Nov	Thu	4		Check yield					

06-Nov	Fri	5		Collect data						
07-Nov	SAT	6			Check yield					
08-Nov	SUN	7			Collect data	Check yield				
09-Nov	Mon	8				Collect data				
10-Nov	Tue	9					Check yield			
11-Nov	We d	1 0					Collect data			
12-Nov	Thu	1 1						Check yield		
13-Nov	Fri	1 2						Collect data	Check yield	
14-Nov	SAT	1 3							Collect data	
15-Nov	SUN	1 4								Check yield
16-Nov	Mon	1 5								Collect data
17-Nov	Tue	1 6	Arrive Maputo							
18-Nov	We d	1 7	Depart Maputo							
19-Nov	Thu	1 8	Arrive UK							
23rd November to 19th December (24 days) - Analysis of detailed score										
4th January to 6th February (30 days) - Yield analysis and final report										

The above programme of visits (Table 3) will be required in order to try and ensure that the trials are carried out properly and that good quality quantitative information is collected. Should Dr. Topper be required to supervise the proposed trial programme, he unfortunately will be unable to undertake all the work. I therefore suggest, that Dr a. Maddison (pathologist and phytosanitation expert) carries out the first two visits and some of the analysis.

In addition to the above programme of visits to Mozambique, it will be necessary to produce a manual so that all technicians will have something to refer to, to help them when in doubt. The manual would cover all aspects of the following:

- C sampling for PMD (both methods of scouting and detailed scoring)
- C *Helopeltis* damage scoring
- C yield data collection
- C spraying/dusting
- C safety
- C sanitation

The GOM scientist and Dr. Maddison could translate the manual into Portuguese.

Three days will also be required for ordering the equipment, consolidating it in the UK and organizing it to accompany the consultants.

2.5 Major Issues to be Resolved in the Near Future

The estimated cost of clearing Nassuruma trial site is approx. \$1,500 (a lot less than the initial estimate of \$7,000); USAID is not in a position to cover this cost, therefore local funding will be necessary to cover this cost. Also, two full time technicians will be required at this site.

It was suggested (by Mr. Camoes and Dr Prasad, Cashew Rehabilitation Project) that the 20 technicians who will supervise the sanitation work should each receive incentive bonuses of \$50 at the end of a successful sanitation period (May) and a further \$50 at the end of successful data collection (October), because this work will be undertaken in addition to their normal programme of work and because they have no transport to easily get to the sites. USAID is not in a position to cover this cost, therefore it needs to be decided if such bonuses are critical to obtaining reliable results and if so, who will pay.

For long term sustainability, it is necessary that a government scientist, who speaks good English, works full time on the trials, learning all aspects of the crop protection programme, following up aspects of the trials and assisting with all aspects of trial work. He/she should be under the direction of the consultant trial manager. The Director of INIA, Mr. Rafael Wyene, suggested that Mr. Matola (agronomist), might be a suitable candidate, in the absence of a person specifically trained in crop protection. The scientist, when not monitoring field trials, should be based in Nampula from about April 1998. All aspects, such as who will pay allowances (and how much), etc. for the driver and GOM scientist while on field trips, should be clearly defined. Sources of funding for fuel and spare parts for the vehicle, etc. need to be clearly defined and accessible.

In order to facilitate monitoring of all the field trials, provision of a good vehicle and driver for use full time on the proposed crop protection trial work is essential.

The sanitation programme should start in January 1998 and before this time it will be necessary to purchase approx. 400 machetes. They are available in Nampula.

Trial protocols should be submitted to the various collaborators (Entreposto, JFS, Cashew Rehabilitation Project, World Vision) for immediate confirmation of their participation under the terms suggested.

The Tanzanian Experience Relating to Crop Protection Issues

Martin *et al* (1997) and Topper *et al* (1997F) have published overview papers describing the general cashew situation prevailing in Tanzania. An overriding problem for cashew research is the very large variation which exists between trees, especially with regard to nut yield, PMD levels, and flowering time (Mead and Martin, 1992, and Neto *et al*, 1994). An extremely important yet very simple finding that affects much of the R& D work in Tanzania is that in a typical farmer smallholding, even with good PMD control, a large proportion of trees, sometimes up to 40 percent, are uneconomic to treat and still produce very low yields. Poor yielding trees remain low yielding over several years; for these reasons selective thinning of such trees is a recommended management practice (Martin and Kasuga, 1995).

Epidemiology of powdery mildew disease has been extensively researched (Shomari, 1996; Shomari and Kennedy, 1997A and B). Methods and keys for scoring PMD have been developed by Nathaniels (1996). While the long-term solutions to pest and disease problems lie with the breeding programme, explained below is the current status of crop protection research in Tanzania.

Chemical control

The application of sulphur dust has been recommended for the control of powdery mildew since the 1980s. Initially the recommendation for controlling powdery mildew on cashew trees was 7 applications of 250g sulphur/tree at 3-week intervals, starting when the trees begin to flush (the development of new reddish leaves). Total sulphur applied per season was 1.75 kg/tree. More recently the recommended rate was reduced to 5 applications of 250g of sulphur dust per tree. Sulphur should only be applied in the early morning when the trees are still wet with dew. Although in many ways sulphur is well suited for controlling PMD in the cashew growing areas of Tanzania, it suffers from one major disadvantage in that after a few years, soil pH declines, leaching of valuable nutrients, increases and in more extreme situations, the level of toxic ions can increase. The length of time taken for measurable effects to occur ranges from 3 to 6 years depending on soil type (Majule *et al*, in press, Ngatunga, 1997). Smith *et al* (1995) found that sulphur deposition on cashew trees was less than 20% of that applied and much lower still if applied when the trees were dry. Further, 3 weeks after application, sulphur deposits on the canopy had declined by 80%.

Boma *et al* (1997A) present the results of a number of trials designed to evaluate the effectiveness of sulphur dust from various sources and water based sulphur formulations. Different dusts have been tested mainly due to cost considerations and water based sulphurs because of their assumed greater efficiency in terms of application (up to nearly 50% of sprays reach the target compared with less than 20% of dusts, Smith *et al*, 1996), and their smaller

particle size, which gives a much greater surface area of sulphur to act on the mildew. In order to improve overall efficiency of controlling mildew with sulphur, dose rates and application frequency have also been researched. The main findings were:

- C water based sulphurs performed better than dusts;
- C there were no significant differences between the sulphur dusts from different sources;
- C the level of mildew control increased with increasing dose rate and application frequency; and
- C there was little scope for reducing the recommendation of 5 applications of 250g of sulphur.

The initial concern over the detrimental long-term effects of sulphur gave rise to a concerted effort to find alternatives to sulphur for controlling PMD. The final objective of this work was to develop an appropriate overall fungicide application strategy, in terms of choice of fungicide, rate of active ingredient, timing and volume of application. The first step was the screening over a 2 year period of a total of 15 fungicides using flushing leaves on seedling plants and then inflorescences on young trees; 6 fungicides were identified for further evaluation (Topper *et al*, 1997A and 1997B).

The screening work was followed by testing the efficacy of 6 fungicides on two large-scale field trials using mature trees while varying dose rate, volume of application, and time of application. The organic fungicides Bayfidan, Anvil, Topas and Folicur were shown to control powdery mildew on cashew more effectively than the currently recommended sulphur dust. There was no significant difference between application volumes of 1 and 5 litres/tree, but 0.5 litres/tree resulted in significantly worse mildew levels. There was no difference between applying fungicides in the early morning and afternoon (Topper *et al*, 1997C).

The final stage was the large scale on-farm testing of a number of mildew control strategies over a 2 year period. This clearly demonstrated that the organic fungicides, Bayfidan and Anvil, controlled powdery mildew on cashew more effectively than the currently recommended sulphur dust, and that this improved control resulted in higher yields. While 250g of 99 percent pure sulphur/tree applied 5 times are needed for effective control, only 10 to 15ml of organic fungicide/tree applied 3 or 4 times/season are needed. Spray volumes as low as 0.75 litres/tree and 1.0 litre/tree are feasible for trees with canopy diameters of <10m and >10m respectively. No significant difference was found between the application of fungicides in the early morning and afternoon, whilst sulphur dust is best applied during a short period in the early morning when the trees are covered in dew. Bayfidan and Anvil were recommended for use in controlling cashew powdery mildew (Topper *et al*, 1997D)

Obviously profitability of using chemical control depends upon the relative cost of inputs (fungicides, blower depreciation, and fuel), the farmgate price of cashew and the yield potential of the trees, all of which will vary from place to place.

Cultural control (phytosanitation) of powdery mildew

Up to 30 percent of smallholder farmers in Tanzania use chemicals to control PMD, which is a very significant number considering the limited resource base of cashew farmers and the fact that credit facilities for such farmers are virtually non-existent. However, that still means that the remaining 70% have no options at their disposal with which to tackle the PMD problem. Because of this and after lengthy discussions as to whether cultural control (phytosanitation) could conceivably work, a programme was instigated to evaluate its potential.

In Tanzania, the *Oidium* epidemic reaches its peak in the dry season when new flushing leaves and flowers are abundant on most trees. The most common site for carry over from one dry season to the next is on shoots and inflorescences inside and under the main canopy. In theory, the removal of these shoots, prior to the main period of leaf flush and inflorescence production, might act to delay the development of the epidemic and assist in the management of the disease. Five researcher managed trial sites were evaluated and when compared with nearby non-sanitized trees, there was “an appreciable advantage for sanitation in every site early in the epidemic, but the differential lessened as the season progressed. Regarding the effects on yield, it is not clear yet whether a delay of a few weeks in disease build up is sufficient alone to improve production, but it may reduce the number of fungicide applications required (Maddison *et al*, 1997).

The potential problems are:

- C expecting the farmers to do something new which they have not seen proven to work;
- C inability to reduce inoculum levels low enough to have a beneficial effect on yield (even though it might have delayed the start of the epidemic);
- C getting a large enough area sanitized to reduce the invasive potential of mildew spores; and
- C getting enough farmers to cooperate in order to do the sanitation well and over a sufficiently large area.

Insect pests

Prior to the increased importance of cashew powdery mildew, sucking insects were the major pest constraint to production, in particular the sucking pests *Helopeltis anacardii* Miller (Heteroptera: Miridae), *H. schoutedeni* Reuter and *Pseudotheraptus wayi* Brown (Hemiptera: Coreidae) a number of authors have emphasized the importance of *Helopeltis* and the need for chemical control (Swaine, (1958) Mutter and Bigger (1961), Northwood and Kayombo (1970)).

The practice of controlling powdery mildew disease by sulphur dusting and organic fungicides has once again resulted in the increased availability of shoots attractive to insect pests, and the sucking pests again pose a serious threat to cashewnut yields, at least in some areas.

Sucking pest leaf damage can take the form of black lesions on petioles or on the leaf midrib or black angular spots on the leaf surface; on stems it appears as an elongate black necrotic area or lesion around the point of entry of the labial stylet into the plant tissue. The damage typically called 'Dieback' involves the withering of the inflorescence or shoot, followed by progressive dieback starting from the tips and advancing downwards to the main floral shoots and leaves; shoots progressively turn brown/black and fruit or new shoot formation is arrested. In very serious cases, the entire tree looks burnt. There was a certain amount of confusion as to the main cause of dieback in Tanzania; one school of thought suggested that fungal pathogens (e.g. *Phomopsis anacardii*) were responsible and another, sucking pests or a combination of both.

Topper *et al* (1997E) described and quantified sucking pest damage on leaves, panicles and fruits and provided preliminary behavioral and developmental observations. Field trials have shown quite clearly that applications of insecticide can prevent the occurrence of black lesions and dieback and increase yields, through the control of sucking pests. If fungi are involved, then it is only as secondary saprophytic colonizers. (However, this does not necessarily mean that under different conditions, fungal pathogens may not cause a different type of dieback). a preliminary insecticide control strategy has been developed (Boma and Topper, 1997).

The control of cashew insect pests is made difficult by the fact that damaging populations are extremely variable from year to year and place to place. *Helopeltis* populations tend to build up on cashew from May/June to September/October, coinciding with the period of leaf flush and panicle development. Under intense feeding pressure, shoots are killed, causing dieback, which results in loss of yield and in some cases, total loss (Boma *et al*, 1997B). The study of *Helopeltis* is further complicated by the fact that it is difficult to sample, as the eggs are injected into the plant tissue so that only 2 small hairs are visible, nymphs and adults are particularly mobile and often hide on the under side of leaves, and adults can also fly off if disturbed.

Pigeon pea, cassava, sorghum and upland rice, four food crops commonly intercropped with cashew, were assessed for their suitability as alternative host crops for *Helopeltis* spp. Only pigeon pea was attractive to *Helopeltis* and intercropping pigeon pea with cashew was found to significantly increase the level of damage sustained by cashew (Topper *et al*, 1997G). This could be a significant factor in Mozambique, since pigeon peas are extensively grown and harvested at the same time as panicle production on cashew.

Severe attacks by sucking pests are particularly pernicious because the farmer could have already spent a considerable sum protecting his trees from mildew disease only to see his yield decimated by insect pests. Seedlings and young trees are particularly prone to insect attack and for a longer part of the year. This is particularly important for young grafted plants where the improved scion material can be killed by *Helopeltis*, leaving only the unimproved rootstock to continue growing.

The biological control of sucking pests has been extensively researched in Tanzania. The presence of the weaver ant, *Oecophylla longinoda* (Hymenoptera: Formicidae) has been shown to have a significant effect on reducing damage and it is possible to assist these predators in colonizing new trees and thereby enhance their capacity for control. a major obstacle to increased expansion are other antagonistic ants, (Stathers, 1997). Peng (1997) has also done similar work in Australia.

The long term objective of the Tanzanian R&D programme is to provide the smallholder farmers with a basket of options to cover all aspects of cashew and the knowledge with which to allow farmers to adapt these technologies to their individual socio-economic and environmental circumstances. The Integrated Cashew Management (ICM) programme was instigated in 1994 to improve the transfer of knowledge and technological skills to a range of farmer types, ensure the relevance of such information, and increase farmer influence on the research agenda. The ICM programme is presently being written-up and will result in the publication of a book entitled “Participatory Research and Extension; a case history of the Integrated Cashew Management programme in Tanzania.”

Appendix 2

Cashew fields visited, observations made and comments regarding future work (all dates are for October 1997)

Nassuruma research station and farmers fields in afternoon (Wednesday 8th)

a large block of local type trees, abandoned for a few years, is available for trial work. a few trees from this block have been selected as “mother trees” for cloning.

The block needs complete cleaning and completion of fire breaks before the next flowering (approx. cost is \$1,500). The trees need re-numbering and the whole block should be mapped. a quick visual yield (arising from the first flowering cycle) estimate should be made of all trees in the block.

Other things seen:

Topworking (2 trees)

The clone collection

Seedling nursery facilities (located across the road from the main sub-station next to a stream)

For details of the breeding, horticultural and seedling multiplication programme, see Dr Prasad's quarterly reports and Prasad and Americo, 1997.

JFS estates at Meserapane and Muchelia (Thursday 9th) Meserapane. Over 10,000 grafted seedlings planted out over the last 2 years, these are a combination of clones from ex-Brazilian material, selected mother trees from Nassuruma research station and locally selected mother trees. All plants are still quite small, PMD levels are quite high but trees are yielding reasonably well. The ex-Brazilian material has PMD and some are quite badly affected. The leaves of many trees have chlorotic areas indicative of a nutrient deficiency. No fungicide or insecticide trials have been undertaken by JFS, although they are growing custard apple plants (*Annona squamosa*) in order to use the leaf extract (leaves boiled up with water) to control *Helopeltis*.

Cotton is widely grown in this area, where the early main season pests are aphids and jassids followed by the bollworms, *Heliothis*, *Pectinophora* and *Earias*. Around 6 insecticide applications are applied each growing season.

JFS was contracted by USAID to produce grafted seedlings and distribute them to the farmers.

Muchelia. The main operations on this estate are cattle rearing, sisal production and cashew. The cashew trees were widely spaced and spread over a large area and had been abandoned for some time. New planting material is being planted in the spaces. The site was not suitable for trial work.

Entrepuesto plantations at Monapo (Friday 10th) a large block of 8 year old trees that runs either side of the main road to Quixaxa was identified as a potential trial site to evaluate chemical control of PMD. The trees are of local origin and 8 years old, they have been described as semi-

dwarf, but apparently this is not the case. The trees had been ring weeded and harvesting had just started. Mildew was widespread, but the presence of a large number of fully developed dried flowers, brown in colour, would suggest that PMD did not develop until sometime after the first flowering. It is this type of information that needs to be collected next flowering season.

A block of 3-year-old Brazilian dwarf trees was also identified as a potential trial site for the evaluation of chemical PMD control and for determination of the effect of mildew on the Brazilian material.

A 2-year-old block of ex-Brazilian material exhibited a number of problems:

Serious *Helopeltis* damage, shoot dieback (possibly with secondary infection) and nut damage

C Complete and partial defoliation of some trees, the leaves dried out, turned brown and dropped off. It would appear that the problem is spreading. Leaf and soil samples were sent to South Africa for analysis but apparently nothing abnormal was found. It is possibly a wilt disease. If the problem continues to spread, then it could become quite serious.

Lepidopterous larvae were observed stripping the lower epidermis off leaves on a few trees.

C Leaves on other trees had black patches, which could have been the result of chemical phytotoxicity.

The nursery and grafting operations were seen.

Mazioteca farm(World Outreach Mission), 8km east of Manapo (Saturday 11th). At this site there are about 800 ha of 25 to 35 year old cashew trees, many trees are anemic looking (lime green colored leaves and low leaf density) with black spindle like panicles with a few dried brown flowers, seemingly unaffected by PMD. PMD was present on many trees. The trees are now too crowded and ideally, alternate rows should be topworked and possibly, legumes grown in the space created to help increase soil fertility, reduce the cost of weeding and provide income to at least cover the cost of these activities. There is a possible 2 ha area for a demonstration plot/validation plot (not statistically analyzable) for 2 organic fungicides. *Oecophylla* was present on some trees

Collaboration potential is good if one good technician is available. The site could provide information on flowering cycle, PMD levels, *Helopeltis* damage, *Oecophylla* and for defining factors for low yield in the absence of PMD.

JFS Geba plantation and cashew factory (Saturday 11th). Large block of local trees (approx. 80ha) visited. Significant *Helopeltis* damage seen on some trees, while on trees colonized by *Oecophylla*, the damage was much less. PMD was widespread. This presents a good site for work on both PMD control and the biological control of *Helopeltis* using the predacious ant, *Oecophylla*. a parasitic weed (*Cassytha spp*) was present on many trees; the whole block is in need of cleaning up, but this is already in the JFS programme.

Work to be done:

Clean and put in regular fire breaks

March/April - Map the position of trees within natural sections (i.e. sections delimited by roads or fire breaks), noting which trees have *Oecophylla*

June/July - PMD scoring and *Helopeltis* damage

Start on *Oecophylla* expansion

A visit round the cashew-processing factory was made; last season it exported 250 tons of processed nuts. Processing of this season's nuts was well underway.

Visit World Vision cashew programme at Muecate (Monday 13th). Trees in the vicinity of homesteads and roads were healthy looking, but affected by PMD. Away from these areas, trees were unhealthy looking, with lime green colored leaves on many trees and low leaf density. Many panicles were thin, with few lateral branches and black in colour. Some had a few flowers which had completed their normal development and then dried out to a brown colour, these appeared to have been unaffected by PMD. Some might have been remaining from the previous flowering season? It is important to determine the factors responsible for such trees. This area was suggested as a possible site for sanitation work, but it was rejected on the grounds that even if PMD was controlled, the trees would still yield very little.

Details of the method of sanitation and practical demonstrations of what should be removed from a number of trees were given.

Visit the Cashew Rehabilitation Project extension activities (Tuesday 14th). An ambitious programme to try and control PMD by cultural methods has been proposed at 20 sites distributed in districts south of Nampula. On this visit to one potential site, the trees were very variable with a range yield potential. Many trees had suckers and flushing under the canopy leaves harboring a lot of mildew.

Details of the method of sanitation and practical demonstrations of what should be removed from a number of trees were given.

Visit World Vision cashew programme at Nacarora (Thursday 16th). Two potential sanitation sites visited; trees at both sites were generally healthy looking in contrast to many of the trees at Muecate. Some trees were yielding in spite of widespread PMD and harvesting was well underway.

Many trees had new flushing leaves and were producing new panicles; for some trees it would appear that the second flushing cycle is more important than the first.

Pigeon pea, a host plant of *Helopeltis*, was seen harboring a large population of black ladybirds. *Oecophylla* were present on some trees and minor *Helopeltis* damage was found.

Collaboration - these 2 sites will be used in the collaborative sanitation trial programme. Details of the method of sanitation and practical demonstrations of what should be removed from a number of trees were given.

Appendix 3

References

- BOMA, F. and TOPPER, C.P. (1997). Evaluation of factors causing dieback syndrome on cashew trees in Tanzania and development of a preliminary control strategy. Proceedings of the Int. Cashew and Coconut conference, Dar Es Salaam, Tanzania. 17th to 20th February, 1997.
- BOMA, F., TOPPER, C.P. and ANTHONY, J. (1997) Evaluation of various sulphur formulations for the control of powdery mildew (*Oidium anacardii* Noack) on cashew in Tanzania. Proceedings of the Int. Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania
- BOMA, F., TOPPER, C.P. and STATHERS, T. (1997) Population dynamics of *Helopeltis* spp. on cashew in southern Tanzania. Proceedings of the Int. Cashew and Coconut conference, Dar Es Salaam, Tanzania. 17th to 20th February, 1997.
- HEINRICHS, E. a. (1997) Development of pest management strategies for cashew in Mozambique. Office of International research and Development, IPM Collaborative Research Program, Virginia Polytechnic Institute and State University, Blacksburg, VA, USA.
- MADDISON, a., BOMA, F., TOPPER, C.P. AND SHOMARI, S. (1997) Sanitation in the management of cashew powdery mildew disease in Tanzania. Proceedings of the Int. Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania
- MAJULE, A.E., TOPPER, C.P. AND NORTCLIFF, S. (1996) The environmental effects of sulphur dusting cashew trees in southern Tanzania. Submitted to *Tropical Agriculture*, 1996.
- MARTIN, P. and KASUGA, L.. (1995) Variation in cashew tree yields in south-east Tanzania and the implication for management of cashew smallholdings. *Trop. Agric. (Trinidad)* **72** (4) 261-268
- MARTIN, P.J., TOPPER, C.P., BASHIRU, R.A., BOMA, F., DE WAAL, D., HARRIES, H., KASUGA, L. J., KATANILA, N., KIKOKA, L.P., LAMBOLL, R., MADDISON, a., MAJULE, A.E., MASAWA, a. E., MILLANZI, K.J., NATHANIELS, N.Q., SHOMARI, S.H., SIJAONA, M., STATHERS, T. (1996). Cashew nut production in Tanzania: Constraints and progress through integrated crop management. *Crop Protection*, **16** (1), 5-14
- MEAD, R. AND MARTIN, a. (1992) Report on farmer participation research, on-farm research and cashew research. Naliendele Agricultural Research Institute, PO Box 509, Mtwara, Tanzania.
- MUTTER, N. E. S. and BIGGER, M. (1961) Cashew. Bulletin 11, Tanganyika Min. of Agric.

NATHANIELS, N. (1994) Major pests and disease problems in cashew in Mozambique. Report to the Danida Plant Protection Project, Ministry of Agriculture, Maputo, Mozambique.

NATHANIELS, N. (1996) Methods, including visual keys for the assessment of cashew powdery mildew (*Oidium anacardii* Noack) severity. Int. J. Of pest Management, **42** (3) 199 – 205.

NETO, V., CALIGARI, P. D. S. AND MEAD, R. (1994) Yield variability of cashew trees in East Africa. Int. Symposium on Plantation Crops (PLACROSYM XI). 30/11 to 3/12, 1994, Calicut, Kerala, India.

NGATUNGA, E. L. (1997) First decade of sulphur dusting in South Eastern Tanzania – implications for soil pH. Proceedings of the Int. Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania

NORTHWOOD, P. J. and KAYOMBO, H. Y. (1970) Cashew production in Tanzania. World Crops **22**, 88-91

PENG, R., CHRISTIAN, K. and GIBB, K. (1997). Bioecology of *Helopeltis pernicialis* (Heteroptera: Miridae) with particular reference to the control efficiency by *Oecophylla smaragdina* (Hymenoptera: Formicidae). Proceedings of the Int. Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania

PRASAD, M. and AMERICO, U. (1997) Genetic upgrading of cashew in Mozambique. Proceedings of the Int. Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania

RAO, Y. P. and NUVUNGA, B. (1993) Report on survey of cashew pests and diseases in Inhambane province, 25 – 30 April, 1994. DSV, Ministry of Agriculture, Maputo, Mozambique.

RAO, Y. P. and UACIQUETE, a. (1993) Report on survey of cashew pests and diseases in Inhambane province, 27 –28 October, 1993. DSV, Ministry of Agriculture, Maputo, Mozambique.

SHOMARI, S. H. (1996). Studies on the biology and epidemiology of *Oidium anacardii* (Noack). The powdery mildew pathogen of cashew. PhD Thesis. University of Birmingham. UK

SHOMARI, S.H and KENNEDY, R. (1997A). Field and laboratory investigations on the development of *Oidium anacardii* in relation to environmental factors. Proceedings of the Int. Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania

SHOMARI, S.H and KENNEDY, R. (1997B) Investigations on factors affecting survival and perennation of *Oidium anacardii*. Proceedings of the Int. Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania

SMITH, D.N; KING, W.J; TOPPER, C.P; BOMA, F; AND COOPER, J.F. (1995) Alternative techniques for the application of sulphur dust for the control of powdery mildew caused by the fungus *Oidium anacardii* in Tanzania. *Crop Protection*, **14**, 7, 555-560.

SMITH D.N., KING W. J., TOPPER, C.P., MHANDO, H., AND COOPER, J.F. (1996). Low volume application of water based organic fungicides to cashew trees for the control of powdery mildew caused by the fungus *Oidium anacardii* in Tanzania. *Crop Protection* in press.

STATHERS, T. E. (1997) Studies on *Oecophylla longinoda* (Hymenoptera: Formicidae) for insect pest management of *Helopeltis* spp. and *Pseudotheraptus wayi* on cashew in Southern Tanzania. Submitted to *Bull. Ent. Res.*

SWAINE, G. (1958). a preliminary note on *Helopeltis* spp damaging cashew in Tanganyika Territory. *Bull. Ent. Res.* **50**, 171-182.

TOPPER, C.P., BOMA, F. and ANTHONY, J. (1997A) Screening trials for the evaluation of fungicides for the control of powdery mildew (*Oidium anacardii* Noack) on cashew in Tanzania. a. Using leaves of seedling plants. Proceedings of the Int. Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania

TOPPER, C.P., BOMA, F. and ANTHONY, J. (1997B) Screening trials for the evaluation of fungicides for the control of powdery mildew (*Oidium anacardii* Noack) on cashew in Tanzania. B. Using inflorescences on medium sized trees. Proceedings of the Int. Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania

TOPPER, C. P., BOMA, F. AND MHANDO, H. (1997C) Evaluation of fungicides for the control of powdery mildew (*Oidium anacardii* Noack) on cashew in Tanzania. a. Fungicide strategy development trials. Proceedings of the International Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania.

TOPPER, C. P., BOMA, F. AND MHANDO, H. (1997D) Evaluation of fungicides for the control of powdery mildew (*Oidium anacardii* Noack) on cashew in Tanzania. B. On-farm testing of fungicide control strategies. Proceedings of the International Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania.

TOPPER, C.P., GRUNSHAW, J., PEARCE, M., BOMA, F., STATHERS, T. and ANTHONY, J. (1997E) Preliminary observations on *Helopeltis* and *Pseudotheraptus* damage to cashew leaves and panicles. Proceedings of the Int. Cashew and Coconut conference, Dar Es Salaam, Tanzania. 17th to 20th February, 1997

TOPPER, C.P., MARTIN, P. J., KATANILA, N., KIKOKA, L., LAMBOLL, R., MASAWA, P.A.L. and SHOMARI S.H. (1997F) The historical and institutional background of the Tanzanian

cashew industry. Proceedings of the Int. Cashew and Coconut Conference, 17th to 21st February, 1997, Dar Es Salaam, Tanzania

TOPPER, C.P., PEARCE, M., GRUNSHAW and ANTHONY, J. (1997G) Preliminary observations on the role of common intercrops in the population dynamics of *Helopeltis* and damage to cashew. Proceedings of the Int. Cashew and Coconut conference, Dar Es Salaam, Tanzania. 17th to 20th February, 1997

3. A SUMMARY OF THE POTENTIAL OF NEW TECHNIQUES FOR BREEDING AND MULTIPLYING CASHEW

The objective of this chapter is to describe in simple terms the work that has been undertaken and is still in progress on genetic fingerprinting, micropropagation and micrografting of cashew, at Wye College (University of London), Wye, Kent TN25 5AH, U.K.

Most of the information presented here results from a visit to Wye College to discuss the on-going work and from recent publications from the scientists working there; it is not intended to be an exhaustive review of the three subjects. The report is presented in three parts:

- Genetic fingerprinting;
- Micropropagation; and
- Micro-grafting.

3.1 Genetic Fingerprinting

The long term objective of a breeding programme is to produce planting material that can give the farmer maximum profit, which in developing agricultural situations, often means crops that can produce reasonably good yields, with minimum inputs (i.e. fertilizers, pesticides, etc.) It follows that in the case of cashew, the released material should be resistant/tolerant to the prevailing major diseases and pests. In East Africa the most important disease is *Oidium anacardii* (powdery mildew disease or PMD) and the most important insect pest is *Helopeltis spp.*

In Tanzania, which has one of the more advanced cashew breeding programmes, this has taken the form of selecting the best trees from research collections (which includes accessions from different parts of the world) and from local farmers' fields. Clones of all promising trees have been evaluated under controlled conditions. Selection so far has been based on morphological characteristics (mainly yield, tree size and nut size) in the absence of inputs like fertilizers and pesticides. a controlled cross pollination programme has recently started to produce new clones (see Appendix 1 to this chapter for the abstract from the paper by Harries *et al* (1997) outlining the proposed crossing programme for Tanzania).

The phenotype (the sum of the characteristics manifested by an organism) can be strongly influenced by the environment and developmental stage/age of the plant. Because the environment obviously varies from one location to another there is a necessity for determining the degree of genotype x environment (G x E) interaction. Basically, this means a clone might perform well in one location, while poorly in another, due to environmental differences. An example of this would be a potentially high yielding clone, but which is susceptible to PMD. In a low mildew environment, it would yield well, but when planted in a high mildew pressure area, the yield would be negligible.

Identifying parental lines to exploit heterosis² and for introducing valuable characters into the cashew breeding programme would be assisted by more reliable information about the level of genetic diversity in gene pools available around the world. If certain characteristics, e.g. PMD resistance, large nut size, dwarf canopy, etc., could be identified or marked genetically, it would then be possible to identify these traits in the different clones in the various germplasm collections. This information could then be helpful in planning a breeding programme with the objective of incorporating these traits into new clones.

In recent years a variety of techniques have been developed which can provide genetical “fingerprints” of populations within a species.

3.1.1 Genetic fingerprinting of cashew and its potential³

Many of the constraints of phenotype-based selection can be alleviated by direct selection of genotypes using biochemical markers based on protein isoenzymes or by more recently available DNA markers, such as Restriction Fragment Length Polymorphisms (RFLPs) and Random Amplified Polymorphic DNA (RAPD). Polymorphisms (distinct kinds of DNA or individuals belonging to one species) found in DNA have introduced a new dimension to crop improvement particularly in the mapping of agronomically important traits. This new technology could be useful for the improvement of plant species such as cashew for which there is no genetic information available.

Various Polymerase Chain Reaction (PCR) based strategies have been described and are used routinely for auditing genetic diversity in plants. Analysis of random amplified polymorphic DNA (RAPD) has been successfully used to assess genetic diversity within several plant groups. The RAPD technique developed by Williams *et al.* (1990) is based on the use of short primers of arbitrary nucleotide sequence in the polymerase chain reaction. Some of the recent examples where this technique has been used to assess genetic diversity include the following: African cassava (Marmey *et al.*, 1994), European and Mediterranean faba bean (Link *et al.*, 1995), oil palm germplasm collected from Africa (Shah *et al.*, 1994) and coffee germplasm (Orozco-Castillo *et al.*, 1994). The study by Mneney *et al.* presents the results of preliminary experiments to explore the possibility of developing Randomly Amplified Polymorphic DNA (RAPD) markers for assessing genetic diversity in cashew. The main objectives of this work are as follows (the study is still ongoing):

1. Preparation of PCR-amplifiable DNA from cashew leaves;

² Heterosis - increased growth vigor in a cross between two genetically different lines, as compared with growth in either of the parental lines.

³ Most of the information in this section originates from Mneney *et al.* (1997) which was published in the Proceedings of the Cashew and Coconut Conference, Dar es Salaam, February, 1997

2. Optimization of PCR reaction conditions to obtain relatively consistent RAPD profiles;
3. Assessment of RAPD polymorphisms in cashew accessions from diverse geographic locations and possibly uncover any geographical pattern of genetic variation in the cashew accessions;
4. Assessment of RAPD polymorphisms in Tanzanian cashew lines; and
5. Recommend alternative procedures for assessing DNA / genetic diversity in cashew, with a view to detecting markers linked to important traits like nut size and disease resistance.

For a general treatise on DNA finger printing, see Weising *et al* (1995)

One of the main aims of this study was to optimize procedures for isolating DNA from cashew leaves, of a high enough quality for PCR and other DNA based analyses (Objective 1). Without this the remainder of the work would obviously have been impossible. However, this was achieved using a CTAB-based procedure that yielded DNA free from contaminating substances like phenolics, polysaccharides and lipoproteins. a mini DNA extraction procedure was also developed that was rapid, involved only a few manipulations, and is therefore well suited to processing large numbers of samples.

As regards the reproducibility of RAPD profiles (Objective 2), RAPD analysis has been described as “a useful and sensitive, yet inherently precarious tool for identification of DNA polymorphisms,” although the RAPD method is appealing as it can be used to analyze any DNA template, does not require radioisotopes, and can be adapted for processing large numbers of samples. However, the RAPD profiles are also known to be influenced by a number of factors. To minimize such errors, many of these factors were standardized initially. For most of the primers tested in this study the RAPD profiles were relatively consistent. However, all PCR reactions were replicated (twice or three times) to confirm the reproducibility of RAPD bands and resolve inconsistent amplification profiles. In this case, the necessity for repeated replication represents wasted resources and makes RAPD analysis of DNA an inefficient tool for a large-scale evaluation project using DNA markers.

PCR analysis of cashew DNA using random-10 mer primers revealed differences in RAPD profiles in cashew accessions derived from different geographic locations e.g. Brazil, Africa and India (Objective 3). Relatively uniform RAPD profiles for the small selection of random 10-mer primers tested suggested a high degree of DNA level similarity within the Tanzanian accessions (Objective 4). This observation will have to be confirmed using more primers to construct a more conclusive picture about the genetic similarity of the different clones. By the time this work is concluded, up to 50 primers will have been evaluated.

There are a number of problem areas with the technique at the moment, including:

1. The large amount of resources consumed to reveal and analyze the RAPD DNA polymorphisms in comparison to potentially more robust marker systems;
2. The low levels of RAPD polymorphisms which have been observed so far; and
3. With such a random approach as embodied in this technique, it will be difficult to know where in the genome such bands are derived from and difficult to convert the observed polymorphisms into robust and reliable markers.

As mentioned before, this work is still on going and many more primers (up to a total of 50) still need to be evaluated, both singly and in combination, and might provide solutions to the above problems. However, it might be necessary to evaluate non-random approaches (see Section 3.3) for assessing genetic diversity, in which case the above work and experience will still be of great value.

3.1.2 Robust, reliable and non-random approaches for assessing genetic diversity

DNA markers must be reliable to be useful for developing DNA-based solutions to assess and exploit genetic diversity. An ideal DNA marker system must be able to resolve questions about the level of genetic diversity in indigenous and exotic cashew germplasm, the selected lines currently being distributed to farmers, and, defining markers linked to economically important characters like nut size, resistance to fungal pathogens etc. RAPD analysis, particularly with some of the problems seen in this study, is likely to have limited uses in such a wide scenario. a dedicated (non-random) PCR-based approach that can query multiple known locations in the genome and detect 'useful' genetic variation will be ideally suited for this goal. Identifying a large number of locations in the genome that correspond to genes and developing markers that detect variation around such sites will be one useful approach. a future strategy might include the construction of Sequence Tagged Site (STS) and Expressed Sequence Tag (EST) libraries of cashew and using genomic locations of ESTs as focal points for assessing DNA variation. One major advantage of such a dedicated approach is the ease with which polymorphisms associated with these markers can be converted into molecular tools and easy to score markers. Together with a high throughput DNA preparation procedure already developed, it should now be possible to envisage a procedure for large scale screening of populations for assessing genetic diversity and to develop marker-assisted breeding systems for cashew.

3.2 Micropropagation

The growing of cashew from seed results in high levels of genotypic and phenotypic variability, the use of grafting techniques overcomes this variability by reproducing the mother plant exactly (although there is still the unknown effect of the rootstock).

The most widely used approach is tip grafting where the only capital investment is a sharp knife and strips (grafting tape) to fix and protect the scion. Rootstock are normally produced by growing individual seeds in polybags. After the seedling reaches a certain height, the top is cut off

and a wedge shaped cut made in the top of the stem into which is fixed the scion material, the tip of which has also been shaped into a wedge in order to exactly fit the rootstock. The union is then bound with grafting tape, which is removed after the union is secure. The whole operation takes approximately 3 months. The success rate varies due to a number of factors, but particularly important is high humidity, hence grafting is often carried out during the rainy season. By the time the grafted plant is ready to be planted out, the rainy season can be well advanced, which under adverse wet season conditions can leave little time for the plant to become established, before the onset of the dry season. The availability of appropriate scion wood can be a limiting factor. Also the influence of the rootstock is unknown.

A number of other techniques are available for multiplying cashew clones, see Behrens, (1996), for an overview of these; a more general treatise on the subject of plant propagation is given by Hartman *et al* (1997).

3.2.1 Background to micropropagation

Cashew (*Anacardium occidentale* L.) is propagated mainly by seeds which results in high levels of genotypic and phenotypic variability (Philip and Unni 1984). Conventional vegetative propagation methods, (e.g. air layering, mound stooling, grafting or cuttings) are not sufficiently rapid. Techniques like micropropagation via multiple axillary branching, micrografting and *in vitro* organogenesis or embryogenesis, offer prospects for faster rejuvenation and multiplication of elite genotypes where this is required, in performing germplasm introduction and evaluation programmes. However, cashew, like other Anacardiaceae, is strongly recalcitrant to *in vitro* culture techniques and only limited successes have been achieved as yet.

Although direct somatic embryogenesis from mature and immature cotyledon sections has been reported (Hegde *et al.* 1990, 1991), a protocol for large scale somatic embryo production for this tree crop has yet to be established. More importantly, the predominant use of zygotic embryo-derived explants with unpredictable genetic potential presented little prospect for their more practical use in rapid clonal propagation. Some progress has been made with application of micropropagation techniques to cashew using microcuttings of seedlings by Lievens *et al.* (1989) and Leva and Falcone (1990).

The main reason is the presence of secondary metabolites which are oxidized after wounding and which cause subsequent browning and necrosis of explants (Das *et al.* 1996). Also, it has been observed by Leva and Falcone (1990) that whilst pale-green calluses exhibited morphogenic activity, brown ones only grew in an unorganized manner. The secondary metabolites of cashew appear to be released mainly from ducts that were observed in the primary phloem elements of all organs. Das *et al* (1996) found that while only 15 percent of cotyledonary nodes survived culture establishment, daily transfer of explants onto fresh medium with activated charcoal for a total period of 7 days increased explant viability to 60 percent and, together with cultivation in darkness for 1 week, survival of explants could be raised further to 90 percent.

3.2.2 The potential of micropropagation

The European Union is funding a collaborative project for the “Development of selection and clonal propagation techniques for multiplication of elite yield and *Anthraxnose* tolerant cashew (*Anacardium occidentale* L.)”. The institutes collaborating on this activity include:

- Unit for Advanced Propagation Systems, Horticulture Section, Wye College, University of London, Wye, Ashford, TN25 5AH, U.K.
- Centro de Investigacao das Ferrugens do Cafeeiro, Instituto de Investigacao Cientifica Tropical, Quinta do Marques, 2780 Oeiras, Portugal.
- Departamento de Fitotecnica e Fitossanidade, CECA, Universidade Federal de Alagoas, Cidade Universitária, 57072-970 Maceio, Brazil.
- Department of Horticulture, Instituts Hassan II, Rabat, Morocco.
- ARI Mikocheni, P.O.Box 6226, Dar Es Salaam, Tanzania.

Most of the information that follows comes from a paper prepared by Mantell *et al* (1997) presented at the February 1997 Dar es Salaam conference on cashew and coconut and a recent Wye College Ph.D. dissertation.

The paragraphs below briefly summarize micropagation methods and results. In order to demonstrate the many factors involved, a brief description of the methods involved are given.

In a typical experiment shoot nodes were collected from 1-month, and 1-, 4 - and 5- year old cashew grown in glasshouses under controlled heat and natural daylight and day length conditions. The 1-month old material consisted of progenies from two dwarf clones improved in Brazil (CP-09, CP-1001) and one Tanzanian (AC4) selection. Explants were surface sterilized. Generally they were incubated on media consisting of either WPM or MS salts with ½-strength macroelements supplemented with thiamine-HCl, inositol and sucrose. The MS medium supplemented with ascorbic acid, glycine and sucrose was used to study the effect of the combination of IAA or GA₃ with one of the following cytokinins, BAP, 2-iP, Kin, Zea, during bud development *in vitro*. The media were solidified by technical agar. Explants were kept at 25°C under 16 h light, 8h dark photoperiod. The percentage bud sprouting, length of shoots and number of nodes per extended axillary shoot, were scored after 4 weeks in culture. For rooting two approaches were tried:

1. Shoots were cultured on medium (pH 5.5) with WPM salts, thiamine-HCl, inositol, sucrose, Technical agar and different levels of IBA (auxin) for 5 days and then on the same medium lacking this auxin; and
2. Well-elongated shoots (with an average of 3 well-formed leaves) were treated during 24h in the dark in IAA or IBA solutions (pH 5.8). After this treatment, shoots were transferred to plain and ½-strength MS salts (pH 5.8) with Morel and Wetmore vitamins, ascorbic acid, glycine, sucrose, Phytigel, where they remained for more than 6 days in darkness, followed by a photoperiod of 12h light/12h dark. Growth

regulators were added.

3.2.3 Micropropagation results

Several factors significantly affected axillary bud sprouting and shoot development. Explants from the two Brazilian selections showed poorer sprouting and elongation than those from the Tanzanian one. Also, in the latter genotype, bud sprouting decreased with plant age while shoot elongation was decreased only slightly. Axillary branching was achieved only with the Tanzanian material. In the following experiments Mantell *et al* focused on this genotype. Media solidified by agar were more suitable for shoot multiplication than liquid media. The use of purified agar resulted in highest bud sprouting, shoot elongation and node formation; moreover, shoots were more vigorous and produced more leaves than those grown on other media. The presence of AC in culture media significantly decreased bud sprouting, particularly in the case of the Tanzanian material. Conversely, shoot elongation was improved by the presence of AC in all three selections evaluated and the shoots produced were invariably more vigorous than those grown on media without AC. Of the five salt compositions tested, MS with ½-strength macroelements proved the most suitable for shoot multiplication. WPM gave satisfactory results also with regard to bud sprouting while White's medium gave the poorest results for all parameters evaluated. No significant differences were found between the lengths of shoots on the five different salt combinations tested. Presence of sucrose in media in increased concentrations slightly decreased bud sprouting. When 20 g/l sucrose was substituted by either equimolar levels of glucose or maltose (but not fructose), significantly more explants formed shoots. The number of nodes per shoot was significantly higher after application of each oligosaccharide while shoot elongation was only slightly improved.

Bud sprouting was significantly influenced by cytokinins. In the case of explants obtained from glasshouse-raised stocks, the cytokinins BAP, Kin and TDZ significantly suppressed bud sprouting while Zea and 2-iP had no suppressive effects. Callus formation was promoted by the highest level of BAP, Zea and Kin whereas callus was induced by TDZ at all concentrations tested. Shoots sprouted on medium with TDZ were hyperhydric and stunted in their growth. Bud sprouting from explants of *in vitro*-produced stock was inhibited by all of the cytokinins tested. Again the cytokinins BAP, Kin and TDZ significantly reduced shoot elongation in sprouted buds from 1-year old stock. By using the cytokinins Kin and Zea, it was possible to produce 6 nodes per main shoot. When used in appropriate concentrations, cytokinins were able to induce multiple axillary branching. Branches grew to lengths of 0.5 cm in 4 weeks and after cutting and further culture of these in the presence of a cytokinin they elongated and produced new laterals. One-year old material exhibited poorer branching ability than that originating from one-month old stock.

The study of the combination of either IAA or GA₃ with one cytokinin (BAP, 2-iP, Kin, Zea), showed that in the presence of GA₃, bud sprouting was strongly dependent on the kind of cytokinin used. The number of leaves/microshoots produced *in vitro* was significantly different within the four cytokinins tested, showing that the addition of 2-iP stimulated while BAP inhibited the production of leaves in the microshoots.

As would be expected, the growth regulator GA supported bud sprouting and shoot elongation; however, the number of nodes per shoot was not affected. Of the three gibberellins tested (GA₃, GA₄ and GA₇), GA₄ was the most promotive for bud sprouting and GA₃ for shoot elongation. *In vitro* microshoots of cashew, produced on high concentrations of GA, however, were weak and hyperhydric and their leaves were much reduced in size. In addition, the microshoots were not able to respond to rooting treatments (see below). Incubation temperatures in the range 19-29°C did not affect bud sprouting, but when they were raised to 35°C, bud sprouting was drastically reduced and profuse callus occurred at the bases of microshoots.

Microshoots formed roots after IBA treatment for 5 days for root induction. The degree of adventitious rooting on cashew microshoots obtained using two comparable root-inducing treatments (IAA or IBA solutions) were not significantly different. After 15 days, roots were already visible emerging from callus tissues produced on the bases of microshoots. The rooting percentages obtained reached 30% in both treatments, and the numbers of main roots varied from 1-12 roots/microshoot and all of these showed similar degrees of elongation and secondary root development. After rooting initiation, microshoots elongated and showed new leaves. Rooted plantlets were weaned and hardened in jiffy-pots containing 1:1 sand/peat mixture, at 26°C and high humidity obtained by covering the plantlets with a plastic bag. The mean survival obtained using this weaning strategy was 100%. One month after weaning, plantlets produced new leaves and at this stage they were transferred to larger pots under open (shaded) glasshouse conditions. After a further two months, plantlets had reached 100-120mm in height and carried approximately 10 leaves ranging from 50-120mm in length.

3.2.4 Micropropagation summary

Several cultural factors affected axillary bud sprouting and shoot development of cashew *in vitro*. Increasing ages of stock plants resulted in drastic decreases in the ability of axillary buds on nodal explants to sprout. Efforts are currently being made to increase the responses of adult phase nodes, by first micrografting adult phase meristems onto seedling rootstocks to obtain partial rejuvenation of elite vegetative buds, before these are subsequently used for micropropagation.

It was concluded that shoot development from axillary buds of cashew probably requires a two-step procedure: the first in which high doses of cytokinin induce well-developed buds followed by a second in which low levels of cytokinin are deployed for shoot elongation. This situation is analogous to the rooting process that consists of several phases characterized by different, often opposite, sensitivities to growth regulators and other active substances (De Klerk 1995).

As small cashew shoots are difficult to root, Mantell *et al* attempted to elongate them first by application of one of three forms of GA. This growth regulator is known to support bud sprouting and shoot elongation in many woody plants including cashew. The positive effect of GA on bud sprouting and shoot elongation was accompanied by later problems with rooting of microshoots. Also preculture or culture of cashew leaf explants on rooting medium supplemented with GA significantly reduced their rooting abilities (Boggetti 1997). Inhibitory effects of GA on root

induction have been documented also for other woody plants.

It was supposed that, since cashew is a tropical plant, culture of shoot nodes at high temperature ($>30^{\circ}\text{C}$) might have improved sprouting of buds and may have favored shoot elongation. However, bud sprouting was strongly suppressed at 35°C although this temperature supported good shoot growth and node development. Again, it seems that a two-step procedure, i.e. a first step in which a lower temperature could be used for induction of bud sprouting, might be followed by a second step, in which higher temperatures are used for obtaining optimal shoot elongation.

Rooting of microshoots derived from nodal explants would appear to be more problematic than those of cotyledonary node origin. This may be simply due to the fact that the latter materials are more juvenile in character. Forty-two percent of microshoots derived from nodal explants of 1-year old stock were able to root following exposure to IBA for 5 days and about 30 percent of microshoots from 4- and 5-year old stock rooted after a 24h treatment in either IAA or IBA. Recently Das *et al.* (1996) increased dramatically the rooting of cotyledonary node-derived microshoots of cashew using *Agrobacterium rhizogenes* transfection. Currently at Wye, they are attempting to use a similar approach (as initiated by Bogetti, 1997) to increase the rooting of shoot node-derived microshoots, using various wild and disarmed strains, with proven root promoting activity, in a model woody perennial *Solanum aviculare*.

In vitro techniques could therefore be used in the future for rapid multiplication and international transfer of elite cashew genotypes. An additional advantage of axillary buds is that they are available in large numbers from single elite adult plants. The research at Wye College has been able to demonstrate that single nodes were able to produce microshoots *in vitro*, some of which had a potential for sustained shoot multiplication in the presence of a strong cytokinin such as BAP.

3.2.5 Costs of setting up a tissue culture laboratory

The estimated costs of setting up a small tissue culture facility capable of producing up to 500,000 units is given in Table 1 (adapted from Hartman *et al.*, 1997) on the next page. Larger scale operations would require more than one of many of these items.

Obviously, operating costs (labor, spare parts, chemicals, etc.) would be in addition to the above costs. a suitable laboratory area would also be required to be set aside for this work, with plumbing, reliable electricity supply, and modern fittings that can be kept clean easily,

Table 1. Estimated start-up costs for a tissue culture facility capable of producing up to 500,000 units. Basic equipment required by most laboratories, costs are per item and presented in 1996 dollars

Item		Approximate cost *
Autoclave		\$8,000
Pressure cooker		450
Laminar flow hood	3 @ 2,500 =	7,500
Glassware		1,500
Test tubes and containers		6,000+
Refrigerator/freezer		750
Water purification system		4,500
Dishwasher		1,000
Dispenser for media		1,000
Chemicals		1,000
Lights and racks for cultures		700
Sterilizer	3 @ 300 =	900
pH meter		450
Balance Top loader		850
Balance Analytical		2,000
Safety equipment		300
Carts	4 @ 350 =	1,400
Electronic stirrer		150
Stir plate/hot plate	2 x 200=	600
Microwave		300
Incubators		3,000
Rotary shakers		1,500
Microscopes		2,000
TOTAL \$		45,850

3.3 Micrografting

A micrografting procedure had not been developed for cashew, although previous work at Wye had demonstrated that it is feasible to micrograft closely related *Anacardiaceae* like pistachio (Abousalim and Mantell, 1992), and that direct shoot and root regeneration can be achieved using the proximal ends of cotyledons (Philip, 1984; Philip and Unni, 1984).

3.3.1 Micrografting methods

Rootstocks used for micrografting studies were obtained from a number of different sources depending on the objectives of the investigations and the location at which experimentation was conducted. Mature cashew nuts were scarified and rigorously surface sterilised. The scarified nuts

were then cultured intact on MS salts medium supplemented with sucrose and agar, to germinate. Once seedlings had reached 5 - 8 cm in height they were removed from jars and decapitated just below the cotyledons. a small angled incision was made on one side of the hypocotyl 8mm below the decapitation point at 45°C to the horizontal plane, using the tip of a sharp fine scalpel blade and the cut opened slightly with a gentle twisting action. a shoot apex prepared immediately before the preparation of the seedling rootstock was then introduced proximal end downwards into the opened incision on the side of the hypocotyl. The seedling rootstock was then reintroduced into its original container to recover and establish the micrograft under the humid conditions. Apical micrografts were carried out by making a vertical incision 2mm in length down the center of the hypocotyl. For evaluation of different sizes of micro-scions, immature seed were harvested from field-grown, healthy mother trees at 5-6 weeks after fertilization (i.e. at the stage of development before nut shells become fully hardened). Nuts were then surface sterilised, a short incision was made along the convex surface of the nut shell and the nut split open to reveal the intact embryo embedded within the kernel, which was removed intact and washed. After washing, the kernel was surface sterilised. The reddish brown testa was then removed and the kernel plus immature embryo cultured on supplemented MS salts medium. For micrografting, seedlings raised from germinated embryos and removed from the jars at 14 - 21d after culturing, were decapitated at 5-8cm up the extended hypocotyl region. Scions were dissected out of shoot buds of field grown mother stock trees of different Tanzanian selections, grown at the Cashew Development Center (CDC) at Mkuranga. Placement of dissected scion buds (of decreasing size ranges: 3-4, 2-3, 1-2, 0.5 - 1.0 and <0.5mm in length) was carried out with the aid of a x10 - 40 magnification range binocular microscope set up inside a laminar flow cabinet.

3.3.2 Results

No significant difference was found in the proportions of graft unions achieved using the side and the apical micrografting methods in preliminary trials at Wye. The micrografting techniques described for pistachio, peach and citrus used seedlings decapitated above the cotyledonary whorl. No information was available in the literature on the effects of micrografting below the whorl (hypocotyl grafting), to avoid the proliferation of axillary buds, which could compete against the developing micro-scions. In the next experiment, epicotyl micrografting was compared to hypocotyl micrografting, using a side graft attachment in both situations. The results indicated that there was no significant difference in graft success levels between the two forms of micrografting. However, there was a measurable and significant difference in the elongation growth rates of each type of micrograft. The hypocotyl grafts grew more strongly than epicotyl ones. By 18 weeks after micrografting, micro-scions on hypocotyl grafts had obtained a mean length of 36mm while those on epicotyl ones had attained only a mean length of 18mm. The effects of scion size micrografted in apical positions on seedling hypocotyls, showed that the larger the micro-scion size, the greater the chance of graft success.

3.3.3 Summary of micrografting

The results of the preliminary work reported in Mantell *et al*, indicate that micrografting of

cashew is a relatively straightforward procedure and that best growth of micro-scions can be obtained with hypocotyls rather than with epicotyls as the stock. The absence of axillary buds in the former probably results in less competition between the developing micro-scion meristem and other buds. Even adult phase micro-scions could elongate following hypocotyl side micrografting and, more significantly, there was no obvious episodic growth present in elongated micro-scions. The more consistent and prolonged vegetative growth in adult micro-scions could now be exploited to great advantage, to supply rejuvenated explants for rapid clonal micropropagation (see Section 2), of elite adult clones, prior to or following their international transfer in the form of micrografts.

This work is on going. It is possible that cashew germplasm introduction programmes aimed at improving genetic resistance in cashew to diseases and pests through clonal selection might now make use of the combined micropropagation and micrografting techniques described above.

3.4 Summary

At Wye College much progress has been made in all three of the above areas of research with a view to improving prospects for multiplication of trees with better production capabilities. It is unfortunate that the RAPD PCR analysis with arbitrary primers has so far proved inefficient and that more work will be necessary in this area before practical results may be achieved. However, for the time being it seems that this is worth pursuing, as it may give very useful results in the final analysis.

Clearly micropropagation requires a reasonably advanced technical base. It is obvious that the work is involved and complex, but can yield viable plants. With a little more research it should be possible to develop a protocol that could result in the mass propagation of elite clones. There is now a team of people working at Wye with good experience in these areas, particularly with reference to cashew.

Appendix 1

A Cashew Breeding Programme for Tanzania

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Abstract

At Naliendele ARI, a breeding programme to generate new cashew clones started in 1996 and is expected to continue for the foreseeable future. Controlled cross pollinations are planned annually and made using standard seed parents in a crossing garden where 101 selected clones are available. The pollen parents are individual trees, from both Tanzanian and overseas accessions, selected from the trials planted at Naliendele. The crosses aim to combine complementary qualities from parents with contrasting characteristics, taking care to prevent inbreeding depression by avoiding parents with a common ancestry. The seed produced is germinated and grafted onto mature seedling rootstocks to appraise, in one or two years, hypersensitivity to powdery mildew, bud vigor and kernel quality. a selection-rejection ratio of 1:20 is proposed for this stage. The selected plants are multiplied by budding or grafting for growing in progeny row trials within existing planted areas at Naliendele. The plants are appraised over three consecutive crop years for growth and vegetative habit, *Helopeltis* tolerance, powdery mildew resistance, with and without chemical control, yield in terms of nut weight and number and quality as percent kernel out-turn. The selection-rejection ratio at this point might be 1:50 and it may be possible to use some selections as pollinators in the crossing garden. The material remaining after the second selection is top-worked for performance testing in on-station and on-farm trials. These trials are also appraised for at least three consecutive crop years under regular farm and plantation management. Some selections may continue to be used as pollinators. Additional selection criteria at this stage includes evaluation by farmers and cashewnut buyers and takes GxE interactions into account. The final selections replace inferior clones - in the seed gardens to improve polyclonal seed production - in the scion gardens to be available for vegetative propagation and - in the crossing garden to become parents for the next cycle of crosses. The new clones will be named for release through the Ministry of Agriculture's Variety Release Committee. General information about them will be available to farmers in a Cashew Clone Catalogue and detailed performance data will be published. Now that the breeding programme has started, new material will become available for testing every year for as long as crossing continues. Unavoidably, the first round of crossing, appraisal, and selection will take up to nine years to complete. Thereafter, new material should become available for release every year.

Appendix 2

Bibliography

- Abousalim a. and S.H. Mantell. (1992). "Micrografting of Pistachio (*Pistacia Vera* cv Manteur)." *Plant Cell Tissue and Organ Culture*, **29**: 3, 231-234.
- Behrens, R. (1996). *Cashew as an Agroforestry Crop: Prospects and Potentials*. Margraf Verlag, PO Box 105, 97985 Weikersheim, Germany.
- Boggetti, Barbara. 1997. "Development of Micropropagation and a Potential Genetic Transformation System for Cashew (*Anacardium Occidentale*l.)". Ph.D. Dissertation, Wye College.
- Das, S. and S.Jha. (1996). "In vitro propagation of cashewnut." *Plant Cell Rep.* 15:615-619.
- Hartmann T. and D.E. Kester, F.T. Davies and R.L. Geneve. (1997). *Plant Propagation: Principles and Practices*. Prentice Hall International, Inc.
- Hedge, M. and M. Kulasekaran, S., Jayasankar, and K. G. Shanmugavelu. (1991). "In vitro embryogenesis in cashew (*Anacardium occidentale* L.)." *Indian Cashew J.* XXI (4): 17-25.
- Hedge, M. and M. Kulasekaran, K.G. Shanmugavelu and S. Jayasankar. (1990). "In vitro culture of cashew seedlings and multiple plantlets from mature cotyledons." *Ind. Cashew J.* XX (2): 19-24.
- Leva, A.R. and A.M. Falcone. (1990)."Propagation and organogenesis *in vitro* of *Anacardium occidentale* L." *Acta Hort.* 280:143-145
- Lievens, C. and M. Pylyser and P. Boxus. (1989). "Clonal propagation of *Anacardium occidentale* by tissue culture." *Fruits* 44:553-557
- Link. W, Dixkens, C., Singh, M., Schwall, M. and Melchinger, a. E. (1995). "Genetic diversity in European and Mediterranean faba bean germplasm revealed by RAPD markers." *Theoretical and Applied Genetics*, **80**: 27-32.
- Mantell, Sinclair H. and Barbara Boggetti, Ana M.S.Bessa, Eurico E.P. Lemos, Abousalim Abdeldadi and Emmarold E. Mneney. 1997. *Micropropagation and Micrografting Methods Suitable for Safe International Transfers of Cashew*. Cashew and Coconut Conference. Dar es Salaam.

- Marmey, .P., Beeching, J. R., Hamon, S. and Charrier, a. (1994). "Evaluation of cassava (*Manihot esculenta Crantz*) germplasm collections using RAPD markers." *Euphytica*, **74**: 203-209.
- Mnene^{1,2,3}, Emmarold E. and Sinclair H. Mantell², Georgios Tsoktouridis^{1,2}, Sawsan Amin¹, Ana M. S. Bessa and Madan Thangavelu¹, (1997), *RAPD-Profiling of Tanzanian Cashew (Anacardium occidentale L.)*
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- Orozco-Castillo, C., Chalmers, K. J., Waugh, R. and Powell, W. (1994). "Detection of genetic diversity and selective gene introgression in coffee using RAPD markers." *Theoretical and Applied Genetics*, **87**: 934-940.
- Philip, V.J. (1984). "*In vitro* organogenesis and plantlet formation in cashew (*Anacardium occidentale L.*)."
Ann Bot 54:149-152
- Philip, V.J. and P.N. Unni. (1984). "*In vitro* propagation of cashew for crop improvement." In: Bhaskara Rao EVV, Khan HH (eds) *Cashew Research and Development*. CPCRI, Kasargod, pp 77-82
- Philip, V.J. and P.N. Unni. (1984). "*In vitro* propagation of cashew for crop improvement." In: Bhaskara Rao EVV, Khan HH (eds) *Cashew Research and Development*. CPCRI, Kasargod, pp 77-82
- Shah, F. H., Rashid. O., Simons, a. J. and Dunsdon, a. (1994). "The utility of RAPD markers for the determination of genetic variation in oil palm (*Elaeis guineensis*)."
Theoretical and Applied Genetics, **89**: 713-718.
- Weising, K., Nybom, H., Wolff, K., and Meyer, W. (1995). *DNA Fingerprinting in Plants and Fungi*. CRC Press Inc.
- Williams, J. G. K., Kubelik, a. R., Livak, K. J., Rafalski, J. a. and Tingey, S. V. (1990). "DNA polymorphism amplified by arbitrary primers are useful as genetic markers. *Nucleic Acids Research*, **18**: 6531-6535.

4. BREEDING AND IMPROVEMENT OF CASHEW IN MOZAMBIQUE

Following the recommendation of Dr. Topper's report, Dr. Peter Caligari visited Mozambique in December 1997. Based on his week long visit, he prepared a report that addressed both concerns with current research, and especially selection and multiplication efforts in Mozambique and outlined future actions to address these concerns. That report is the basis for this chapter.

4.1. Introduction

This chapter is based on a consultancy carried out by Dr. Peter Caligari of Reading University in December 1997. His principal assignments were to address:

- C the immediate issues of selection and multiplication of cashew material suitable for extensive exploitation and the methods to be adopted; and
- C the longer potential to establish a breeding programme for cashew.

The visit was made in a time period which restricted its scope to covering Maputo, Ricatla and Nampula Province, but this was regarded as being the main cashew production area and representative of the situation in Mozambique.

The consultancy was carried out in the company of Mr Julio Cuamba.

4.1.1 Mozambique at present

The rise and decline of cashew production is already well documented and the causes reported. a recent report visit by Dr. Clive Topper (Topper, 1997) also summarizes the present position with regard to constraints on production. He highlights the major constraints in relation to biotic factors as being powdery mildew disease, with sucking pests providing additional problems. This accords well with the experience in Tanzania. Another factor that also needs to be highlighted is the age of the material currently growing. There has been little new planting of cashew material (until recently) for many years. This means that the basic stock of trees for commercial exploitation are old and mainly past their optimal production age. There are different estimates of the production potential of cashew trees over time from establishment, but our own recent analyses of long term data from East Africa, including Ricatla, shows that in practice the optimal production is reached between 10 to 12 years after planting (Neto and Caligari, 1998) after which production falls, sometimes dramatically. Clearly this will be influenced by factors such as the spacing between trees, the nutrient status of the soil⁴, the management, the genotypes used, etc. Nevertheless, the implication must be that most of the trees that might be exploited in

4 This consultancy did not uncover any reports of soil analysis being undertaken or proposed in relation to rehabilitating cashew production. This seems important and necessary and hopefully is being addressed elsewhere. If not the author recommends an initial brief review/survey be undertaken.

Mozambique are likely to be past their optimum production. This will have been further exacerbated by the fact that many of the trees were neglected for a long period with the subsequent deleterious effects on their condition. Certainly the yields being obtained are extremely low.

4.1.2 Background

The breeding of cashew in strict terms involves the crossing of chosen genotypes, growing out the progeny, collecting data over years and sites, multiplying selected types, and then exploiting these and/or using them for re-crossing. This is, by definition, a process that takes a considerable number of years in cashew (Ohler, 1979; Harries, 1998) as in other tree crops or even in crop species with an annual cycle (such as potatoes - Caligari, 1992). The risk of multiplying unsuitable genotypes to occupy large areas of land, with this only to become apparent after a period of time, cannot be over-emphasized. Nevertheless the potential long term advantages of producing genotypes with particularly appropriate combinations of characters and with adaptation to the environment in which they are to be exploited are high. a number of factors must be considered:

1. The background knowledge of genetics and breeding in cashew is still restricted (Masawe, 1994); even the natural pollinating mechanism and frequency of out-crossing is uncertain (Masawe and Caligari, 1998);
2. The lag period while a breeding programme *sensu stricto* was established would be unacceptable to remedy Mozambique's present problems;
3. The ideotype of cashew tree needed for the most appropriate sustainable exploitation under various farming systems is still unclear; and
4. The expertise to establish and carry out a breeding programme in Mozambique would need to be established.

This leads to the conclusion that the only viable immediate prospect is to improve the programmes of selection and propagation so that, in the short term, they can provide the necessary improvement of the basic exploitable germplasm in Mozambique. But just as clearly, if Mozambique is to sustain its cashew production, it must set in place a breeding programme as soon as possible in order to be better placed at the first future opportunity.

To carry out any selection and propagation programme, the first requirement is a range of trees showing variable and genetically determined characters (i.e. variable germplasm). There are various possible sources of germplasm among which selection could be practiced. In broad terms:

1. Existing genotypes, and derived progenies, being grown and harvested within the country;
2. Existing genotypes, and derived progenies, in collections within the country;
3. Existing genotypes, and derived progenies, being grown, or in collections, within the close geographic area i.e. East Africa, mainly Tanzania, but also potentially Kenya; and

4. More "exotic" genotypes, and derived progenies, from other parts of the world - principally Brazil.

The prospects of exploiting these different sources need to be considered from a number of standpoints, including relevant existing experience:

Existing genotypes being grown and harvested within the country. In many ways this is an attractive proposition in that the cashew trees are already available close to, or in, the areas that need to be rehabilitated. They are "easily" available and have survived in the location for a period of years and so have some proven adaptability. The question that must be asked is whether they have the inherent characters that are required of any new planting material? Another question that arises, as a subsidiary of this, is how to assess the inherent potential (genotype) of any material when faced simply with its visual appearance and performance (phenotype)? This question is central to the strategy that needs to be adopted and will depend on the character in question and the method of assessment used.

Existing genotypes in collections within the country. Again a potentially attractive proposition, if suitable material can be identified, because of its availability and access. Again, as with the above, the question of it containing suitable variability is vital.

Existing genotypes being grown, or in collections, within the geographic area ie East Africa, so mainly Tanzania but also potentially Kenya. After the use of "local" material, the next obvious possibility is to examine material available in similar and close geographic regions - in other words within East Africa. The proximity of the Tanzanian breeding programme at Naliendele is clearly a prospect that needs to be viewed - in terms of possible germplasm and in view of their recent experiences.

More "exotic" genotypes from other parts of the world - principally Brazil. Because cashew is an introduced species to Africa there is an obvious logic when contemplating variability in germplasm to consider the "Center of Origin" of cashew - which is Brazil. They have a range of material and have been exploiting it commercially. However, the extensive efforts in India to successfully exploit cashew should not be forgotten.

4.1.3 Assessment of germplasm - general

In brief, the characters (or traits) that are important to optimize the production of any crop are usually numerous and complex. In addition, the ones that are important will depend on a variety of factors such as: (1) the system and management under which they are to be grown and maintained; (2) the agronomic practices to be imposed, and (3) the use for which the crop is being grown. The other basic complication is the genetic control of the expression of any of these characters. As an over-simplification, characters are either controlled by a few genes with clearly observable effect (Major genes) or controlled by many genes (polygenes) with indiscernible individual effects and further "camouflaged" by their variable response to their environmental conditions - thus giving continuous variation. The former are relatively easy to deal with but tend

to be uncommon among the characters that are of principal interest (or in the case of disease and pest resistance have been shown to be undesirable because of their disastrous lack of sustained effectiveness).

The basic underlying principles that are appropriate to selection in cashew are broadly similar to those in any other crop species. Cashew, along with other tree crops, has the problem of generation cycles being lengthy. The general conclusion from the formal research (where it has been looked at in formal detail) shows that selection on the basis of single plants is effective for major gene controlled characters but not for those which display continuous variation. With the latter the need for replicated, randomized trials with allowance for judging the potential macro-environmental effects is well established. In other words the need to assess the relative levels of Genotype by Environment interactions is obvious and this needs to encompass both micro- and macro-environmental variables (many of which will always be undefined) (Bos & Caligari, 1995).

To put this in a simple equation:

$$\text{Phenotype (observed)} = \text{Genotype} + \text{Environment} + (\text{Genotype} \times \text{Environment})$$

a simple assessment based on observable phenotype is often very misleading.

4.1.4 Assessment of germplasm - cashew in East Africa

There are a number of aspects relevant to the particular case of cashew and the difficulties that have been faced in practice in relation to the characters that are of most interest. It is generally accepted that, in the context of East Africa, the major breeding objectives are overall yield of nuts, nut quality, powdery mildew tolerance/resistance and, as a slight subsidiary, sucking pest tolerance/resistance.

There is no evidence that suitable major genes exist that give desirable levels of expression of these characters, and so they must be handled as appropriate for quantitative characters (Masawe, 1994).

There is direct evidence of the problems of handling yield assessment in cashew; indeed it is the subject of a recently completed PhD thesis (Neto, 1997). The problem, stated simply, is that when trials are carried out it is extremely difficult to show significant differences in yield between genotypes or clones. This is a reflection of an extremely high "error/environmental" variance pertaining in the trials. This makes selection in trials or the development of crop protection strategies extremely difficult and the outcome uncertain.

As far as mildew resistance/tolerance is concerned, the position is also problematic. a procedure of identifying local individual trees, similar to that being practiced at present in Mozambique, was undertaken in Tanzania over a number of years (Masawe, Cundall & Caligari, 1998). Local trees over a wide area were identified, with the help of local farmers, that appeared

to be resistant to powdery mildew. The trees were inspected, marked and observed over several years. Those trees that retained an acceptable phenotype (i.e. appeared to be resistant/tolerant) were cloned/grafted and trialled in the Naliendele Agricultural Research Station. They were included with other material of various origins that came from the germplasm collection held in Nachingwea, Tanzania. The genotypes from the farmers fields performed disappointingly with only 2 out of 41 yielding more than the control (and neither significantly so). Thus, there was no indication of the expected effect that selection (both natural and farmers driven) was giving rise to identifiable genotypes (rather than just phenotypes) that were resistant/tolerant in terms at least of yield. (As an aside, the trial did identify the fact that one clone, AC4, did give progenies which showed a higher than expected number of resistant/tolerant genotypes and so is potentially of interest for further breeding).

4.2 Assessment of Resources Currently Available in Mozambique

There has been a successful development of the technologies needed for cloning (grafting onto seedlings and/or top-working) any material that is identified as of interest. This has tremendous application in terms of multiplying material for trials, breeding or exploitation. The first consideration must be to exploit cloning to provide a system to reliably test material for its genetic potential (as at Manapo) and hence avoid the spurious mis-identification by phenotype of individuals.

The potential use of Brazilian dwarf material is obvious, at least in a "plantation" system. If the correct material is selected it can be fully productive within about 2 years of planting, it can be planted on a higher density pattern (noting the correct orientation of rows), it allows much simpler management, husbandry and spraying when appropriate. It also can be selected more readily for a "bolder" nut which is clearly advantageous in the world market of processed whole nuts. However, it must be noted that it is still unproven on a large scale in Mozambique and its longer-term potential to perform, show tolerance to pests and diseases will only come with time (thus its selection and multiplication should continue but with continued monitoring). The local material should not be dismissed and its testing needs to continue alongside the Brazilian - in breeding terms it may well represent a very important source of other characteristics. Similarly the germplasm such as that available in Tanzania should be considered seriously and included in trials alongside other material.

The germplasm available from Brazil is somewhat restricted and its wider availability may need some negotiation, but it basically seems to be becoming available in Mozambique generally and so could be used as a basis for selection.

The selection of material for propagation and multiplication must be taken much more seriously and carried out with more rigor. The risks of inappropriate genotypes being multiplied cannot be emphasized too strongly - if nothing else the detrimental psychological effect felt at all levels if all the effort involved only produces trees with performances no better than that at present, is obvious. The potential to try to develop an appreciation and market for the cashew apple is also something that should be taken into account (particularly as material being multiplied

and established is now taking place an opportunity exists for developing a more dual exploitation potential). This needs to be considered now even if it is only likely to be exploited in the years to come since breeding and propagation are always limited in their potential to respond to market demands on a short time-scale.

It is clear that there is effectively no breeding program currently being undertaken for cashew in Mozambique. The present activities are clearly concentrated on selection (much of it almost certainly ineffective) and propagation. This is understandable with the need for short-term progress to be made and breeding itself always needs a reasonable timescale and a consistency over time. Thus if cashew is to continue to be important in Mozambique, efforts must be put in place to establish a breeding programme sooner rather than later.

The sections that follow provide a brief summary and analysis of various sources of genetic material in Mozambique.

4.2.1 Ricatla

Established in 1969, the germplasm collection was gathered from an extensive search over the whole of Mozambique. Thus, it represents a large sample of germplasm that was present in Mozambique at that time. Unfortunately, it was laid out in a systematic manner in such a way that many of the environmental variables are confounded with source of origin. There is also no replication other than within plots. Another problem with this site is that it is really on the extreme edge of the cashew growing region and so its suitability to give representative growth of trees must be questioned (see Neto, 1998, for further details).

Mr. Inicio Armindo is currently taking care of the station on a day-to-day basis and reports to Mr Madola of INIA. Mr Armindo has been in post since 1984 and so knows the material well and is a valuable source of knowledge. He guided us round the collection and showed us a number of small experiments with grafting, particularly involving dwarf cashew material of Brazilian origin (Mr Madola was unfortunately absent at the time of the visit).

The site is unfortunately not well suited for cashew material. The material is now mostly "past its best" and the layout never was particularly well considered for statistical analyses of any data collected. It is important to decide the worth of the germplasm collection and then consider re-establishing (by grafting) worthwhile material at a more suitable site, with a carefully considered design for trials or conservation blocks. The usefulness of the material might be judged to some extent by analyzing the potentially extensive data collected annually since the 1970's. There are strong arguments for taking all the material if it is to remain as a representative germplasm collection. Unfortunately much of this data has been lost and the data sets now held in Reading are probably (apart from some very recent data) the most complete available - these could be exploited to help identify a conservation strategy and possibly start to identify promising clones for further breeding trials and crossing. No formal records of powdery mildew resistance were kept but no attempt was made to control mildew and so the yield figures will reflect performance in the presence of the disease (and, at least partially, reflect tolerance/resistance).

The experiments set up and in progress are on such a small scale and without proper design so as to be uninformative, and nothing will be lost if they are stopped.

The other material, such as the "Fan Trial/Density experiment" has been overtaken by the growth of the bush and is past its useful life. a check should be made as to the existence and state of the data collected on this trial and any conclusions drawn - the trial itself is of no further value.

4.2.2 Nassuruma⁵

The site is well situated in relation to a major cashew growing area, although it is not conveniently sited for general access or external facilities. Most of the "interesting" material present has been planted in the last two years under the energetic influence of Dr Prasad with the encouragement of his Team Leader, Mr Rui Ribeiro. There is a collection of material planted somewhat earlier, but this appears not to have been satisfactorily labeled and its exact identification, mostly from Ricatla, is uncertain. There is an adjacent nursery which has been established very logically near a local water source and equipped with locally available components. This is used to produce "grafted seedlings".

There have been tremendous developments in the last couple of years and the further development of the various grafting techniques to ensure their practical applicability is noteworthy. In addition, there seems to have been a very useful networking of the training and know-how to District and Village level.

There is some dwarf material of Brazilian origin and this is being used as a source of "mother trees" for some of the grafting work and experiments.

There were a few rather small-scale, unreplicated trials to test some of the material and its suitability for use as "mother trees". The work on developing the grafting techniques and spreading them to a village level provides optimism for such an approach and demonstrates its technical feasibility. But the question arises as to whether this is a reasonable approach for local small-scale farmers as opposed to identifying parental material to give superior seeds and seedlings for direct planting.

A practical "systems" review should be undertaken into the best possible approach at the different farming levels. The grafted material gives the potential for production of uniform stands of cashew (i.e. clones), the ability to Top-Work existing trees to re-establish existing plantings and the potential to "spread" new "types" (e.g. dwarf trees) rather quickly. The disadvantages are that this does create large blocks of identical genotype with its concomitant risk (all respond the same, and if a disease or pest establishes, it does so on a large scale). Seed material is genetically less uniform, which can be an advantage (as noted above), but also may give variable performance. This latter is not necessarily a problem for small-scale farmers who simply hand-harvest when appropriate or in the slightly longer term since it is possible to select for parent trees which give rather uniform progenies, (as achieved in potatoes [van Hest, 1994] in relation to their phenotype

⁵ The author's assessment of Nassaruma has been controversial. For a response, please refer to Appendix 1 to this chapter.

(as opposed to their genotype)). The answer is almost certainly a mixture of the two possibilities. The potential to Top-Work will provide a useful and rapid way of rehabilitating plantings that are not too old (up to 10-15 years old). The potential to graft carefully selected genotypes onto seedlings or as Top-Worked material also opens the possibility of being able to produce designed seed orchards to produce seeds of a more controlled constitution given the rate of out-crossing in cashew (Masawe & Caligari, 1998).

The selection of "mother trees" at Nassaruma is based on very limited data, and appears totally inadequate for forming the foundation of an extensive propagation programme. The selection of individual local trees on the basis of simple observations of their phenotype is ineffective and potentially very damaging (as already noted). The use of Brazilian material may provide a source of much greater powdery mildew resistance and this is of great interest, but its value is clearly unproven.

Again, the use of small-scale, rather informal trials to select potential material is not recommended. The low incidence of powdery mildew in Brazil holds the prospect of their material having better resistance than the small sub-sample of germplasm that probably forms much of the East Africa plantings, but the inherent resistance of this material cannot be assumed. The greater incidence in East Africa could be connected with different alternative hosts, environmental conditions, or "races" of the pathogen, etc. In any case the Brazilian material is certainly NOT uniformly more resistant (see notes on Monapo). The testing in relatively simple but well designed trials, using the grafting techniques to produce replication, should be carried out with some urgency to empirically identify the genotypes which are relatively more tolerant/resistant (as carried out in Naliende, Tanzania - the Variety Improvement Trial - Masawe, *et al.*, 1998). On the other hand, claims that Mozambique has identified genotypes that are "better" in terms of mildew resistance than AC4 (of Sri Lankan origin) of Naliende are precipitous. One would certainly hope that better material exists both in terms of disease and pest resistance as well as quality, but it has yet to be proven. It is interesting to note that extensive data from the Nachingwea Collection in Tanzania has generally identified cashew genotypes of "Asian" origin. They had a rather better yield tolerance to mildew than other material in that collection (this did not include Brazilian material) (Neto, 1998).

The increased recognition of nut size as a quality character of some importance to Mozambique is certainly appropriate. Some of the small nut size, at present a feature of Mozambican nuts, can be attributed the generally old and poor state of many of the trees; however, it is also fairly clear that most of the genotypes represent rather undeveloped genotypes as far as nut size is concerned. Given its importance in marketing cashew it clearly warrants attention being paid to it in breeding and propagation activities. It might be noted that the relation of nut size to kernel size is by no means absolute.

It has been suggested that this site be developed as a center for technology training and development. It has certainly been used very successfully and in a short time to give commendable results in terms of propagation techniques (the identification of "mother trees" and the methodology involved has already been noted as more worrying). It might be noted that it will be important to identify any important features to be specifically watched for when these techniques are being used

at the different levels - for instance the depth of the plant growing container is more important than its width, if root damage is to be avoided. However, there is no reason to automatically pick this site for such a center. Its disadvantages include the fact that:

1. It is somewhat distant from any reasonable center for access;
2. Its facilities and services are limited (e.g .phone, electricity, accommodation, etc); and
3. It has few features which could not easily be produced elsewhere.

Other sites, such as the INIA Station on the outskirts on Nampula, retain the regional location and have many of the facilities needed.

4.2.3 Muecate

The site provides a good example of how effectively the technology of grafting cashew has been dispersed to other levels. The site is one based on practical reality at the local level and provides a very promising basis for various "extension" activities with a number of crops. The opportunity to see some of the selected "mother trees" and talk with the local farmers was welcome and indicative of the truly interactive philosophy involved and which Mr Eliezer Carmago is following.

The potential exists to exploit the local germplasm, if appropriate, and pass it directly back to the community. New material, or types, (e.g. dwarf types, almost exclusively ex Brazil) could also be introduced in this way. The selection of the "mother trees" is based on the "Nassuruma philosophy" and clearly is similarly likely to be based on a false assumption about genotype and phenotype. It is also clear that although dwarf material has many attractive features (e.g. easy maintenance, spraying, closer spacing, etc.) it may not be the most suitable for small-scale farmers, given the likely growth of other vegetation and the activities of animals, the potential to intercrop, etc.

The selection process for "mother trees" must be made more vigorous. The potential to clone trees means that any tree can be multiplied on a modest scale into properly designed "trials." This fact must be the basis for justifying the inputs and energy underlying this basic scheme. The potential to actually employ dwarf genotypes under different farming practices needs further investigation.

4.2.4 Namialo

There is material planted here which is basically in two blocks. One block derives from Brazilian dwarf material and the other from local germplasm. All is grafted and up to 2 years old. Unfortunately, there are no records or labels for this material and so its value is greatly reduced. There is an effective nursery for growing seedlings and grafting, and this is being used to provide Mr Joao De Carvalho of JFS with planting material.

The value of the material and any data is very limited because of the lack of information. Missing material is now being replaced with other material as available. The site can only really be viewed as providing a basic comparison of dwarf and local material and so in research or breeding terms is of limited value. It might be considered, if appropriate, for use in being adapted

for disease\pest control trials or for extension experiments.

4.2.5 Monapo

The site and material are carefully maintained with obvious attention to the essential details which provide the basis needed to carry out the research and breeding successfully. Mr Paulo De Carvalho had been involved with cashew in Brazil and has established plantings and schemes based on this useful background. The material being considered was both local and ex-Brazil (being seed progenies from exploited clones in Brazil). The trees were planted in careful rows correctly orientated for sunlight (Masawe *et al.*, 1997) and with clear records and labels. The nursery was well sited and considered, and allowed suitable multiplication practices to be achieved.

The seed material was planted in progeny rows with individual trees identified. Data were collected on a regular basis from every tree and recorded. This formed the basis of the initial selection of potentially interesting individual trees. Once selected (60 trees so far) these were then used to provide scions for modest trials at 2 sites, comprising 10 clonal trees per site - again data were carefully recorded on these. From the data and observation on these clones the material was then reselected for potential exploitation. When a clone was selected, scions were collected from the original individual tree and used to graft in Top-working or to produce grafted-seedling. The identification of material was continued at this stage, an important feature as it would allow a continued and widening basis for continually assessing the material of originating from different trees or origins.

The basic scheme in place under Paulo de Carvalho has a sensible and logical basis both in theory and in practice. The simple but thoughtful organization and care taken provide information that can be used to build up an evermore informative picture of the various material and its potential. This basic scheme might be used as a model to help evaluate the potential of both local and "exotic" germplasm. I would suggest some modest changes and additions to a "model" system, but the basic philosophy seems sound.

Another sign of the organized nature of the whole system at this site was the useful printed leaflets (in Portuguese) giving information that covered the essential features of the techniques involved and thus could back up any practical training.

Without necessarily endorsing all the details of all the leaflets, they clearly provide a potential model set of information that (if permission was given) could be developed further for more general use. There was clearly a variable degree of powdery mildew symptoms in the planted progenies derived directly from Brazilian clones. Indeed Mr Carvalho was undertaking active selection within these for the expression of resistance.

Any thought of the Brazilian derived material being necessarily resistant to powdery mildew is mistaken.

4.3 Summary

In Mozambique the achievements in developing propagation techniques, training local people in their application and the development of appropriate facilities have been commendable. However, the material for cloning is mainly being selected on the basis of inadequate information, often from single trees. This is not a reliable approach to selection except in the case of a few specific characters (like fruit colour and possibly dwarf habit) in other words, generally those characteristics that are controlled by single major gene differences. Other characters are subject to considerable environmental variation (and typically genotype by environment interactions) and it has been shown in many species (including specifically in cashew) that such selection is ineffective, including for powdery mildew resistance.

The cloning being carried out at present is therefore worrying in terms of its potential future implications (e.g. mass propagation of mildew susceptible trees of a uniform genotype) and its psychological impact (in terms of likely response from those involved to the time and effort involved leading to trees which are not materially better). a more logical and structured approach to selection is essential.

An approach with similarities in philosophy to those employed at Entrepосто (Monapo) is essential. This selection programme is based on a multi-stage approach. Although there is initial selection of individual trees (either from fairly large created segregating populations or from local material) which may or may not be particularly effective, the selections are then modestly propagated to give trials at two sites. On the basis of these trials the material is more closely selected. The careful labeling and recording protocols are also essential. This would provide the minimum requirement for the initial phase of a selection and propagation programme and is urgently required.

Thus a major component of the short-term requirements can be met by simple incorporation of more formal protocols which specify a set of properly designed trials. These would be interposed between the stage of having individual trees and a decision to multiply these for exploitation. The implementation of this would be facilitated by a short (one week) training course in appropriate statistics, design, record keeping, trial management as applied to a breeding context could provide a reasonable basis for upgrading these aspects.

A study or a Workshop is needed to define the ideotype for trees that are to be exploited under different farming systems (the main one is the applicability of dwarf trees). Also it is necessary to undertake a rapid appraisal of the applicability of using grafted clonal material as opposed to seedlings under different farming systems and in different areas. This should also include clear recommendations as to when "top-working" on existing plantings is a realistic and a viable alternative to replanting.

In general, a much more structured view must be taken of breeding cashew in Mozambique. Effectively none is being undertaken at present. To have a long term future, cashew genotypes need to be produced by crossing and selection to produce the appropriate genotypes that are

adapted to Mozambique's conditions. This requires suitably trained personnel.

There must be a review and rationalization of the information on cashew material (germplasm) in Mozambique. The need for a physical rationalization of the germplasm is also essential. The acquisition of further germplasm is a further requirement.

An issue that needs addressing is the very extensive data that should exist from the collection in Ricatla. From the evidence available it appears as though much of this has been lost and the best copy of the existing data, in a useable condition, is in Reading. This data should be made as complete as possible and the results exploited as a matter of urgency.

Two possible courses of action emerge as possibilities for supporting the cashew sector in the short-term, a small workshop to help integrate the ideas and approaches and also to clarify issues such as those raised in term of ideotype etc, ending in agreed written protocols would be useful. The second is for an "institute", to be established in an appropriate area of the country. This would help address the aspects mentioned above of a rational bank of germplasm, and because of the requirement for more coordinated testing means such a "structure" is needed urgently - it would also form the basis for a much needed breeding programme for cashew which at present does not exist in Mozambique.

4.4 Recommendations

As the basis for providing the foundations for the development of a longer-term strategy to improve cashew breeding in Mozambique, The AMIS II Team makes the following recommendations:

1. Develop the framework of an "accepted" protocol to screen and select genotypes with an approach based on a philosophy similar to that already employed at Entrepuesto (Monapo) .

Such a selection programme is based on a multi-stage approach. Although there may be an initial selection of individual trees (either from large segregating populations or from local material) this should only be on the basis of characters with proven repeatability. These "selections" should then be modestly propagated to give trials at a minimum of two sites. The sites should be chosen to represent contrasting conditions under which it is intended to grow the resulting trees. On the basis of these trials the material can then be more closely selected. The next stage is to start multiplication BUT to keep records of the plantings and employ at least basic trial design principles to allow further data to be collected and fed-back into the selection process, leading to further refinements in the selection recommendations.

It is also obvious that the same material can also be cloned by Top-Working to allow an additional facet to the selection. In other words, the initially identified material can be Top-Worked into existing plantings where pest and disease pressures will help provide suitable pressures for identifying appropriately resistant/tolerant material - again a valid trial design is needed. The disease pressure can also be artificially strengthened as soon as time and facilities permit (e.g. Masawe, Cundall and Caligari, 1998).

It cannot be stressed too strongly that careful labelling and recording processes are also essential components of the overall protocol. This procedure will ensure that all selections are based on real performances and that mistakes, the consequences of which can be extremely detrimental, are avoided. This needs to be considered in terms of laying out field trials, labelling trees, producing and keeping accurate field plans, recording data, checking data collection, analysing data and applying selection criteria (which must be backed up by physical inspection of the trees).

It is essential that "best practice" as developed and agreed must be fully documented, distributed to suitable centres, kept updated, etc.

These are then the minimum requirements for the initial phase of a selection and propagation programme. They need to be implemented urgently. They would also form the basis for then identifying suitable parents and carrying out crossing programmes to ensure future viable cashew production in Mozambique.

In summary:

- i. Single Trees - Select, if at all, on the basis of reliable characters.
- ii. Clone "Selected" trees - Use scions to produce grafted seedlings and/or Top-Work to give a designed trials capable of statistical analyses: minimum of 5 trees x 2 replicates x 2 sites.
- iii. Data and Selection - Collect data on the above material for at least 2 years in "good" production for all appropriate characters. Either use one of the trial sites or better establish disease and pest screening trials where the pressure from these is high or manipulated to be high (as per Naliendele). Select best material.
- iv. Further cloning - Depending on circumstance further multiply by cloning and use in commercial settings but with suitable trial design and with records and labelling maintained and checked. Collect modest data at regular intervals to monitor on-going performance.
- v. Crossing Programme - Use the above scheme to start a crossing and breeding programme. This would then provide material to stage i. It should be noted that at this stage it would be possible to further reduce the ineffectiveness of individual tree selection by using progeny data (Brown & Caligari, 1986a,b, 1989; Brown *et al*, 1988; Masawe, 1994).

To establish the incorporation of the protocols for selection, which specify a set of properly designed trials, a short (one week) training course is suggested in appropriate statistics, design, record keeping, trial management as applied to a breeding context to provide a reasonable basis for upgrading these aspects. Those who should attend would be those responsible for setting the framework for selection and multiplication. Ideally this should lead to an agreed written protocol for "best practice".

In the longer term, however, it is vital that Mozambique has trained people who have the necessary background and expertise to develop and integrate such approaches into the practical on-going programmes.

2. Conduct studies to define the ideotype for trees that are to be exploited under different farming systems

Studies are needed to define the ideotype for trees that are to be exploited under different farming systems (the main question is the applicability of dwarf trees). The potential of dwarf trees for precocious flowering, (therefore early returns after establishment), and ease of management and control, seem well suited to plantations, but other features of the dwarf trees (e.g. height above the other vegetation, potential invasion of parasitic weeds) may be problematic for small scale farmers. This potential conflict needs clarifying. The genetic base of the Brazilian dwarf material also needs checking since its origin is almost certainly from one genotype, but how many sexual generations ago? How much outcrossing has given rise to the present genotypes and in what background? How suitable are the background genotypes, in the longer term, for Mozambique's conditions? How much should they be intercrossed with local germplasm?

On the basis of these recommendations, decisions about material to be propagated into these systems can be made and the emphasis of any breeding work defined more logically.

3. Assess the conditions under which planting clones or seedlings is appropriate.

Make a realistic assessment of where and under what farming systems planting clones as opposed to seedlings will be appropriate in practical reality. This should also include clear recommendations as to when Top-Working on existing plantings is a realistic and viable alternative to replanting. There are a number of considerations here including the time scales involved. There effectively needs to be a short-term and longer-term policy, which will not be identical, but should be compatible. At present there are virtually no proven clones or progenies or the means of reproducing them on a sufficient scale to give adequate planting material immediately. This can be partially taken care of (with lessening compromise in terms of guaranteed performance) by the exploitation of cloning techniques to both trial and multiply genotypes. The most important aspect is to carry out recommendations given above. On the basis of this an important decision will be needed about any restraint to be placed on the exploitation of a narrow genetic base (e.g. how much should a single clone be multiplied?). It should be noted again, that a check needs to be made of what the genetic base is for the Brazilian dwarf material: is

it really a very restricted base or does it represent considerable out-crossing from the original mutant? The question of distribution of monoclonal crop stands and genotype diversity is a crucial one if cloning of "elite" genotypes is to become widespread - this really requires a greater knowledge of the pest and disease structure and epidemiology, but work in other crops might be used to give some ideas on this. The problem with trying to give a quick answer is that it is not really only the number of clones that might be sensibly used and with what pattern, but what is the genetic diversity of the clones.

4. Analyze the data collected Ricatla to determine whether it gives reliable identification of any material that can be exploited.

Analyze as soon as possible the data collected for so long from Ricatla and decide whether it gives reliable identification of any material that can be exploited. The casual observation of phenotypes carried out there at present for the basis of multiplying individual trees is far from satisfactory.

A very extensive set of data should exist from the collection in Ricatla. As far as can be judged, however, much of this has been lost although (through the good offices of the Secretary of State for Cashew) the team managed to rescue copies of some of it a number of years ago. Since then the team had extensively processed it into a useable condition in Reading. It is far from complete, but has been entered, sorted and "cleaned" by Mr Victor Neto who used some of it as a part of his submission of his thesis (to be examined in January 1998). It would seem sensible to recheck for the existence of any more of the past records, update the Reading database with the most recent year's results, and use it to extract whatever information was readily available, particularly in relation to identifying any promising accessions. Mr Neto's database and knowledge of the data could be usefully exploited to give a quick and informed view as to what can be gained from this data set and any likely material identified.

5. Structured approach to breeding cashew in Mozambique.

In general, a much more structured view must be taken of breeding cashew in Mozambique. Effectively none is being undertaken at present. Indeed there is no coordinated selection being undertaken. To have a long term future, cashew genotypes need to be produced by crossing and selection to produce the future genotypes that are adapted to Mozambique's conditions, while fitting the criteria needed for sustainable production at the same time as giving "best fit" to marketing requirements. This implies the need to have breeders with a sound training in the underlying science of breeding and genetics as well as practical knowledge of cashew. Such training would need to be at least to the level of a Master's degree. I suggest that there is need to consider a minimum of 2 breeders initially but with a plan to quickly add a third - the need for continuity is essential. Thus, aiming for a "senior breeder" and 2 active associates. Clearly, to optimise the position would require that at least the senior breeder is at a PhD level.

6. Careful consideration of cashew germplasm.

The important resource of cashew material (germplasm) must be carefully considered. There is an extensive collection of local material in Ricatla (as noted in 6.4), but this is of ageing material and needs replanting (at a more appropriate site). It could possibly be used as a scion garden⁶. Although there are some genotypes maintained at different sites. It could possibly be used the only other noteworthy organised material is in Monapo (although I do not know how generally this might be available in a wider sense), where a well maintained set of Brazilian derived progenies are planted. A systematic appraisal of existing data (as noted under 6.4) and then more specific trailing of available material is needed. The further acquisition of germplasm is also important. A coordinated programme should be organised using local sites and existing facilities, but with an agreed design and layout.

7. Formation of an Institute with specific focus on cashew.

The suggestion of an Institute with a specific focus on cashew should be considered seriously and quickly. There are clearly numerous issues to be taken into account beyond the scope of this consultancy. But in terms of breeding it is imperative to have a stable and consistent input if it is to succeed, if for no other reason than the time-scales involved in breeding. There is also the question of a germplasm collection since this forms the heart of any breeding programme. The Ricatla collection is probably now inadequate and so a new planting, using grafting, could be set up at a new Institute - where very careful labelling, recording and data handling and analysis would be a major and initial priority.

The initial screening of material could be organised from the institute and crossing work initiated. Depending on the basis of the institute the material from this could then be made available to interested parties - preferably with arrangements about the use of trials or data recording built into the early stages of multiplication and dispersal. This could then be based on an agreed charging system which takes into account the financial status of the recipients and their role in funding the institute.

The siting of such an institute, in terms of breeding work, would need:

- a. Access to nearby land for germplasm collection
- b. Accessibility for moving material to and from site
- c. Access to facilities and services (water - irrigation and laboratory; electricity; storage; machinery up keep and maintenance, etc.)
- d. Reasonable access for national travel, to help ensure contact to all growing areas, processors, international commerce, collaborators, etc.)
- e. Access to suitable land for breeder's trials
- f. Reasonable contact with growers and possibilities of collaborative trials
- g. Access to laboratories for various tests

⁶ Plants maintained in high density planting as small trees/shrubs to provide material to graft onto root stocks. The material is not planted for assessment or trials but simply for maintenance of germplasm.

- h. Seed storage facilities
- i. Suitable screen-houses
- j. Adjacent colleagues in crop protection, agronomy, extension, etc.

It would also need:

- k. Freedom from short-term decisions and changes in emphasis, but with flexibility to respond to changing practical demands. This implies a sympathetic governing body with representation from all levels of the growers, processors, marketing as well as researchers and extensionists, but also with modest international representation to encourage an objective baseline with an outward looking approach.
- l. An appropriate funding base.

Clearly these matters are outside the direct terms of the consultancy, but the long term nature of any breeding programme, the need for a stable basis on which to carry out such a programme, the need to develop reliable recording and database and the necessity to safeguard Mozambique's genetic diversity in cashew means that such this issue is highly relevant to any future planning for breeding and improvement.

8. The potential of new techniques - particularly biotechnology and molecular biology.

New techniques are providing exciting possibilities and opening up new ways of approaching areas such as breeding and propagation. However, they should not be seen as providing an immediate and practical solution for Mozambique. The current situation will mostly be tackled by the application of more "traditional" approaches. Nevertheless, it is important to start considering now the future, sustainable development and exploitation of cashew. This must include consideration of these techniques (reference should also be made to the Chapter 2, by Clive Topper on New Techniques).

- i. The potential for tissue culture systems has yet to be explored in Mozambique. Such work has been initiated elsewhere in the world but is not well developed for cashew generally. It certainly does not hold the prospect of being a universal solution to problems, and it is unlikely to be suitable for direct multiplication for field grown plantings (its costs and technical complexity are likely to rule this out except for specific purposes such as breeding blocks, etc). It does, however, offer some future potential breeding techniques: (1) to provide a disease free nuclear stock storage system; (2) a route for international exchange of germplasm with reduced health risks; and (3) an initial rapid disease free multiplication system for elite genotypes. This is clearly an aspect that should be developed, but would seem to be ideally suitable at this stage as a collaborative venture rather than solely a Mozambique project; perhaps at least East African.

- ii. The potential for molecular biology in terms of molecular markers and genetic transformation is growing and opening what were previously almost unthought of possibilities. Molecular markers allow the unique identification of genotypes as well as giving measures of genetic diversity and the potential for "trait-tagging"⁷. Some preliminary work has been done with cashew, but further development and the use of more suitable systems (such as those developed at Reading for cocoa germplasm and currently being expanded to oil palm and cedars) need to occur. Indeed, work has already been planned to be carried out collaboratively between Reading and Naliendele. Transformation is another relevant technology, particularly when we consider characters such as pest and disease resistance as well as quality. This work still is finding its "true level" even with the crops of more world-wide significance. It is likely to be technology with very powerful applications in a rather specified set of circumstances. Again, it is important to start considering an initial building of expertise and knowledge to be able to exploit the techniques at a slightly later stage, but be able to judge and apply its benefits when appropriate.

This area of development would clearly be something to consider in the light of discussions about an institute (see 7, above). The desirability of developing, perhaps after an initial phase, should be borne in mind in terms of planning. The possibilities of collaborative activities must also be an attractive possibility and make funding possibilities more widely attractive (e.g. EU grant applications).

9. Training

The training requirements to allow the breeding and propagation work in Tanzania to progress satisfactorily are initially modest. The most obvious areas of deficiency at present are:

- i. Increase the awareness, at the higher levels of supplying the expertise in the techniques of propagation and mother tree selection, of selection protocols and their theory and basic aspects of appropriate trial designs, as needed in the scheme given under 6.1.
- ii. Some breeders with enough genetical, statistical and experimental design background to help put a more rational approach to basic trials, their analysis and interpretation. Clearly an overall appreciation of breeding would be required and a Master's level training is suggested.

⁷ The possibility of being able to identify and follow sections of DNA that have particular effects on traits of specific interest - thus allowing a much easier and more rapid assessment and/or selection of characters.

- iii. Somebody with genetic and breeding expertise at the both the theoretical and practical level. Such a person needs to be aware of quantitative genetics, selection theory, applied statistics and cashew biology. They really need to start putting in place a more defined overview of a breeding strategy. It is suggested that a researcher with appropriate interests be identified who has, or should be given PhD⁸ training in a relevant topic area.

8 It should be noted that there is potential at Universities such as Reading for "split-PhD programmes" where the main research is carried out in the student's own country. Basically, they attend the chosen University for initial increase in background skills and knowledge, experiment planning etc, return to own country for actual research activity and then finally return to the University to complete their thesis. This means relevant research can be carried out with direct relevance to the student's country.

Appendix 1

Response of Dr. M.V.R. Prasad, Cashew Rehabilitation Project, Nampula Province

As noted in the main text, Dr. Caligari's original report, which serves as the basis of chapter 3 of this deliverable, stirred some controversy in Mozambique. The pages that follow comprise a response to Dr. Caligari which disputes some of his findings and conclusions. It was prepared by Dr. MV R. Prasad of the Cashew Rehabilitation Project, Nampula Province.

Readers will have to make their own judgements about the validity of the original findings and conclusions and Dr. Prasad's response. The debate on the subject of how best to develop a longer-term strategy to improve cashew breeding in Mozambique does not end here. Rather, it is the hope of the AMIS II Team that the Cashew Working Group will further the debate in which Drs. Caligari and Prasad have enjoined as the basis for a more detailed dialogue on how best to address this critical issue.

CASHEW REHABILITATION PROJECT
NAMPULA PROVINCE

OBSERVATIONS ON COMMENTS

MADE BY PROFESSOR PETER D.S. CALIGARI
ON HIS VISIT TO NASSURUMA ON 02 - 12 - 1997

M.V.R. Prasad
BSc. (AG), M.Sc., Ph.D., F.I.S.G.
JANUARY/1998

Ref.: Breeding And Improvement Of Cashew In Mozambique
Report On Visit To Mozambique - 29th November To 7th December 1997
by Professor Peter D.S. Caligari, BSC Ph. D. DSC. Cbiol. FI Biol FRSA.

OBSERVATIONS ON COMMENTS
MADE BY PROFESSOR PETER D.S. CALIGARI
ON HIS VISIT TO NASSURUMA ON 02 - 12 - 1997

The visit of Prof. Caligari to Nassuruma was very brief lasting around 45 to 50 minutes during which a summary of activities was presented to him. The terms of reference of his visit were not communicated to me . I was only told that he would be interested in seeing new cashew germplasm. As the available germplasm collection at Nassuruma is not clonal in nature, but represents seed progenies showing wide segregations, I preferred to show him some new clonal materials (which Dr. Caligari referred to as unreplicated trials) which I thought could be of some interest.

The methodology adopted for selection of mother trees was not discussed. Dr. Caligari also did not elicit any details regarding the above subject.

In view of the shortage of time only a (1) few new clonal selections of some interest, (2) two top-worked cashew trees and (3) the work on vegetative propagation of cashew at the nursery located away from the center, were shown to him and this took all the time available.

Dr. Caligari asked two questions. The first question was if the soft-wood grafting was a proven technique of vegetative propagation. The second was about the possible instability of a somatic variant I showed him.

The work on cashew improvement of this project is spread out at three different locations viz., (1) Nassuruma, (2) “Posto Agronómico” and (3) “Viveiro Monapo.” In view of the shortage of time it was not possible to visit the other two locations.

Now, I am really at a loss to understand as to how he reached an erroneous conclusion that the selection of “mother trees” was based on inadequate data and simple observations of their phenotype, which is ineffective and potentially damaging !

I would have appreciated had Dr. Caligari commented that the selection methodology was not discussed and as such should be looked into. The consequences of misgivings that the comments of this nature would provoke cannot be over emphasised !

(1) Selection of Mother Trees of Cashew

Now I take this opportunity to explain as to what is the basis of selection of mother cashew trees at Nassuruma. We have two kinds of cashew materials. I.e. (A) selections emanating from the originally introduced segregating seed progenies of five Brazilian cashew varieties consisting of 306 trees grown at Posto Agronómico of INIA and Viveiro Monapo, and (B) Selections made from out of local cashew tree populations in Nampula Province. We have also introduced some material from Ricatla and Gaza province which will not be discussed here.

The above selections made for attributes such as branching and flowering (percent of flowering shoots), nut size, resistance or tolerance to powdery mildew and nut yield.

A. In the case of the material of Brazilian origin thirty single plants were selected initially, following which (a) the seed progenies of these selections at the rate of 20 plants per selected tree fitted in two replications (10x2) were planted in an area of 7.2 hectares in the front zone of Nassuruma station out side the entrance. (b) Also a smaller trial of the clonal progenies of the above selections consisting of six bud-grafts per each selection fitted in two replications (3x2) was grown in another area of 1.5 hectares. Based on the data collected from the above two trials, as well as the performance of the original mother trees in the subsequent years, only FOURTEEN selections were finally retained, rejecting the rest.

Now we possess (i) five years' data in respect of the original mother trees and (ii) four years' data in respect of their seed and clonal progenies, which in my opinion is adequate enough to arrive at proper judgement about the selections we have made. Another set of 12 selections made out of the Brazilian material from the germplasm bank in 1995 - 96 are still being observed and analysed.

B. Regarding the selections of the local cashew trees, those were not chosen in the recent past; but were selected during 1984 - 85 by the staff of the Provincial Cashew Service.

Initially more than 150 trees were selected for higher nut yield in different districts viz., Mogovolas, Mogincual, Moma, Angoche, Nampula and Meconta. The yield data available in respect of the selected trees for all the years with effect from 1986-87 were analysed. The Cashew Project did distribute ungrafted seedlings of these selected plants to farmers during the earlier years starting from 1989 - 90 to 1994 - 95. Now one can see these seedling orchards of farmers even today.

As a part of the germplasm collection at Nassuruma, 450 seedlings of the 75 of the above selected trees were grown in two replications in an area of 5.5 hectares. The data obtained from this trial with effect from 1994, as well as the data from the time tested original mother trees all these years and the seedling orchards arising out of their progenies in villages were useful in re-evaluating them for the above mentioned attributes and arriving at appropriate decisions with regard to the utility of these trees. All the trees exhibiting inconsistent performance were rejected. Now we have also organised a comprehensive replicated trial involving clones of local Mozambican selections as well as the exotic materials.

The clones of selections emanating from the seed progenies of Brazilian dwarf material have not yet been distributed to the farmers, but are being evaluated in the new clonal orchards installed at every Pilot Nursery. The promising material shall be distributed in the year 1998-99.

A comprehensive paper is being prepared incorporating all the available data on this topic of selection of elite cashew material.

Therefore it is not correct to assume that our approach was not reliable and that selection was

based on simple visual parameters.

We are aware of the value of progeny evaluation and testing of the material over seasons in view of the high genotype x environment interactions exhibited by a constellation of characters of agronomic value. We are also aware of the dangers of propagating inadequately tested material.

The selections that are being multiplied at present are well characterised, backed by a sound data base and as such there is no danger of susceptible material being propagated on the mass scale as suspected by Dr. Caligari.

It is necessary to note that we are vested with the mandate of meeting the short term goals of the Cashew Rehabilitation Project in terms of making available grafted plants of superior quality to the farming community.

In this context, the technique of 'soft-wood grafting' has proved very useful as well as efficient in achieving the rapid multiplication of proven cashew selections.

We have also started a programme of hybridization involving appropriate parental combinations from out of the above selections and at present we have some hybrid material too. Our endeavour is to lay a foundation for long term programme of cashew breeding through a wide array of new selections which could not only form a source of new germplasm, but offer new parental material. The technique of top-working is being used as a tool to achieve rapid and full phenotypic expression of the hybrids as well as the original selections.

(2) Clones Vs. Seedlings

This has been very well debated and tested in cashew. Early efforts in distributing the seedlings of the proven elite plants did not enhance the production and as such the only viable strategy available to achieve higher levels of cashew production is through clones of genetically superior cashew varieties (Rao *et al* 1993). Knowing advantages of clones as against seedlings, even the small farmers would prefer to grow only grafted plants as is evident from the Indian experience and as has been observed by us now in the Project area in Nampula province.

Then, how can we over come the disadvantages and risks of creating large blocks of identical genotypes ?

The answer is not in advocating planting of ungrafted seedlings; but to distribute clones/grafted plants of diverse productive genotypes which could be grown together in large blocks, thereby offering an insurance against the out break of disease or pest epidemics. This is the extension of the concept of varietal blends in annual crops (Prasad, 1979; Prasad and Reddy, 1992), when the considerations of product quality are not very important. It would be of added advantage if these genetically different varieties could be phenotypically uniform.

(3) Disease Resistant Material

We never stated that all the Brazilian derived material is necessarily resistant to powdery mildew. We come across highly mildew susceptible material also in the Brazilian collection.

The only statement I made was that the few selections exhibiting resistance to powdery mildew are of Brazilian origin. It certainly does not mean that the genotypes originating from other parts of the world can not be resistant ! We have introduced some germplasm of Asian origin. Some selections made out of this material (which are still under observation) are indicating promise for resistance/tolerance to powdery mildew.

Also, we never claimed to have identified genotypes that are better in terms of mildew resistance than AC4 of Naliendale. Despite the promising performance of few selections showing resistance at Nassuruma, we shall not make such a statement without adequately testing by comparing these two kinds of materials.

(4) Nassuruma Centre

It is not for me to suggest if Nassuruma Cashew Centre should be a national centre of technology and training. The decision in this regard rests with the appropriate authorities.

However, I would confirm the observation of Dr. Caligari that it has been possible to come out with useful programme of work and results in a short time at Nassuruma. From the above, it is clear that a base is being created for future cashew improvement and breeding programmes in spite of shortage and non availability of some facilities.

References:

Prasad, M.V.R. (1973)

Some considerations on plant types for dry land agricultura.

Annals of Arid zone, 3 e 4: 125-134.

Prasad, M.V.R. and Reddy, B.N. (1992)

Ad-vances in Oil Seeds Production under dryland situations in SUSTAINABLE DEVELOPMENT OF DRYLAND AGRICULTURE IN INDIA. Scientific Publishers
Jodhpur (India): 334pp.

Rao, E.V.V.B., Swamy K.R.M., Yadukumar, N. And Sixit, S. (1993)

Cashew Production Technology.

National Research Centre for Cashew, Indian Council of Agricultural Research,
Pultur (India): 37pp.

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Nampula, January 30th, 1998

Appendix 2

Bibliography

- Bos, I and Caligari, P.D.S. 1995. *Selection Methods in Plant Breeding*. Chapman and Hall. 0 412 55330 9) pp x + 347.
- Brown, J. and Caligari, P.D.S. 1986a. The efficiency of seedling selection for yield and yield components in a potato breeding programme. *Ziet. fur Pflanzenz.*, 96, 53-62.
- Brown, J. and Caligari, P.D.S. 1986b. Cross prediction methods in potato breeding programmes. *Proc. of Eucarpia (Biometrics Section)*, Birmingham, 1986, pp. 348-355.
- Brown, J., Caligari, P.D.S., Dale, M.F.B., Swan, G.E.L. and Mackay, G.R., 1988. The use of cross prediction methods in a practical potato breeding programme. *Theor. Appl. Genet.*, 76, 33-38.
- Brown, J. and Caligari, P.D.S., 1989. Cross prediction in a potato breeding programme by evaluation of parental material. *Theor. Appl. Genet.*, 77, 246-252.
- Caligari, P.D.S., 1991. Breeding new varieties. In *The Potato Crop* (Ed P.M. Harris), Chapman and Hall, London, 335-372.
- Harries, H.C., Kusolwa, P.M., Millanzi, K.J. and Masawe, P.A.L. (1998). a cashew breeding programme for Tanzania. *Proc. Int Cashew and Coconut Conf.*, Dar es Salaam, Tanzania. 17-21 February, 1997.
- Masawe, P.A.L. (1994). *Aspects of Breeding and Selecting Improved Cashew Genotypes (Anacardium occidentale L)*. PhD Thesis, The University of Reading.
- Masawe, P.A.L., Cundall, E.P. and Caligari, P.D.S. (1996) Distribution of flower sex-types between clones and sides of cashew tree canopies in Tanzania. *Annals of Botany*, 78, 553-558.
- Masawe, P a. L. and Caligari, P.D.S. (1997) Investigation of cashew apple colour and out-crossing in cashew (*Anacardium occidentale* L). *Proc. Int Cashew and Coconut Conf.*, Dar es Salaam, Tanzania. 17-21 February, 1997.
- Masawe, P a. L., Cundall, E.P and Caligari, P.D.S. (1997) Performance in South-Eastern Tanzania of local cashew germplasm and selected clones. *Proc. Int Cashew and Coconut Conf.*, Dar es Salaam, Tanzania. 17-21 February, 1997.
- Neto, V and Caligari, P.D.S. (1997) The effect of variability on cashew yield trials. *Proc. Int Cashew and Coconut Conf.*, Dar es Salaam, Tanzania. 17-21 February, 1997.
- Ohler, J.G. (1979). *Cashew*. Koninklijk Inst voor de Tropen, Amsterdam. pp260.
- van Hest, P a a M. (1994). Production of potatoes through true botanic seed. *PhD Thesis, The University of Reading*.

5. CONCLUSIONS AND RECOMMENDATIONS: NEXT STEPS

The work that the AMIS II Project has carried out in Mozambique to date has prepared the foundation upon which to develop a comprehensive and detailed cashew production rehabilitation strategy. For this process to go forward, it is now crucial that the Cashew Working Group exert its authority and take a more directive role in the process. Since the initial AMIS II consultancies to investigate and analyze production issues, there have been a number of positive developments that indicate the timing may be right for the Cashew Working Group to mobilize the diverse stakeholders with an interest in improving the country's ability to produce a greater quantity of higher quality nuts.

The single most encouraging development may be the creation of a small, informal group of scientists and technicians in Nampula Province who are interested in production issues. The group includes representatives from the region's major private sector cashew producers, Grupo Entrepuesto and Grupo JFS, the African Development Bank-funded Cashew Rehabilitation Project - Nampula, World Vision International, and the provincial level representatives of the Ministry of Agriculture and Fisheries (MAP) and the National Institute for Agricultural Research (INIA). The group, many of whose members are partners in the crop protection trials described in Chapter 1, meets periodically to discuss production related issues.

This report recommends that the coordinator and secretariat of the Cashew Working Group rely heavily upon this informal group to guide its decisions in planning the next steps to develop a more extensive cashew production strategy. As employees of the organizations which are working most closely with the AMIS II Team to carry out the crop protection trials, these are the individuals who will take a lead role in disseminating the lessons learned from this effort. In addition, they are the practitioners who are best placed and most knowledgeable to suggest to INIA where it should focus the limited resources it has to devote to cashew research, so that the money spent has the greatest impact.

In addition, this group should take a lead role in developing the longer-term portion of the production rehabilitation strategy. The members of this group form the core of the stakeholders who should participate actively in discussions with outside experts to figure out the most appropriate ways for Mozambique to improve the selection of the genetic material that is propagated and distributed to the country's farmers. The members of the group have the collective capacity to review proposed terms of reference for expatriate consultants and the members of the group should serve as the counterparts for such consultants.